A METHOD FOR DETERMINING GENETIC AFFILIATION, SUBSTRUCTURE AND GENE FLOW WITHIN HUMAN POPULATIONS

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CROSS-REFERENCE

[0001] This application claims the benefit of U.S. Provisional Application No. 06/245,355, filed November 1, 2000, which application is incorporated herein by reference.

STATEMENT REGARDING FEDERALLY SPONSORED RESEARCH

[0002] This invention was made with government support under grant nos. GM55273 and GM 28428 awarded by the NIH. The government may have certain rights in this invention.

FIELD OF THE INVENTION

[0003] The present invention relates to nucleic acid polymorphisms and their methods of use in, for example, determination of paternity and forensics.

BACKGROUND OF THE INVENTION

[0004] The science of genetics has taken a keen interest in the identification of human individuals and genetic relationships between individuals. The genome of an individual is unique to that individual, and can be used for identification purposes, e.g., testing for paternity and/or forensic testing (e.g. to identify an individual in the context of post-mortem identification or in the criminal justice system). Procedures have been developed which are based on identification and characterization of changes in an individual's DNA, referred to as DNA polymorphisms, where such changes are due to nucleotide substitution, insertion, or deletion within the chains of DNAs.

[0005] In forensics, for example, there is an interest in polymorphisms for identification purposes. Techniques have been developed to compare homologous segments of DNA to determine if the segments are identical or if they differ in one or more nucleotides. Practical applications of these techniques relate to fields other than forensic medicine, for example, genetic disease diagnosis and human genome mapping.

The most accurate and informative way to compare DNA segments requires a method which provides the complete nucleotide sequence for each DNA segment. Particular techniques have been developed for determining actual sequences in order to study mutation in human genes. See, for example, Proc. Natl. Acad. Sci. U.S.A. 85, 544-548 (1988) and Nature 330, 384-386 (1987). However, because of the extensive amounts of time and high costs to determine, interpret, and compare sequence information, presently it is not practical to use extensive sequencing for compare more than just a few DNA segments.

[0007] A frequently used technique for screening for DNA polymorphisms arising from mutations consist of digesting the DNA strand with restriction endonucleases and analyzing the resulting fragments by means of Southern blots. See Am. J. Hum.Genet. p32, 314-331 (1980) or Sci. Am. 258, 40-48 (1988). Since mutations often occur randomly they may affect the recognition sequence of the endonuclease and preclude the enzymatic cleavage at that site. Restriction fragment length polymorphism mappings (RFLPS) are based on changes at the restriction site. They are accurate but not very informative (PIC) 0.3). The major problem with RFLPs is the inability of a test to detect changes that do not affect cleavage with a restriction endonuclease. In addition, the methods used to detect RFLPs are very labor intensive and expensive, especially the techniques which includes Southern blot analysis.

[0008] Another technique for detecting specific mutations in particular DNA segment involves hybridizing DNA segments which are being analyzed with a complementary, labeled oligonucleotide probe. See Nucl. Acids Res. 9, 879-894 (1981). Since DNA duplexes containing even a single base pair mismatch exhibit high thermal instability, the differential melting temperature can be used to

distinguish target DNAs that are perfectly complimentary to the probe from target DNAs that only differ by a single nucleotide. See, *e.g.*, U.S. Pat. No. 4,683,194. Further, subtle genetic differences among related individuals regarding nucleotides which are substituted in the DNA chains are difficult to detect. VNTR's or Jeffrey's probes are very informative but labor intensive, in distinction to microsatellites which are equally informative PCR based tests.

[0009] Short tandem repeat (STR) polymorphisms are commonly used in DNA identification, either as adjuncts to other genetic tests, or as stand-alone tests. Typically, when STRs are used for human identification, they are amplified in groups of three to four loci (multiplex amplification). Generally, the resulting amplified fragments are analyzed by polyacrylamide gel electrophoresis. Polymorphisms are thus typed according to size by comparing to similarly labeled known external standards or differently labeled internal standards. U.S. Pat. No. 5,364,759 describes the genus of simple tandem repeats as well as a DNA typing method employing the simple tandem repeats and PCR amplification of the loci. Fragments are analyzed by differential labeling of the products.

[0010]A critical parameter in DNA typing is the power of exclusion for the system. Power of exclusion is the ability of a test to exclude a falsely accused individual based on the individual's genetic characteristics. The commonly used STR multiplexes have exclusion probabilities in the range of 85% to 91%. This compares unfavorably with restriction fragment length polymorphic loci (RFLP loci), which often provide an equivalent power with just one locus. STR testing batteries which include greater numbers of lower power systems are more susceptible to this problem than are RFLP testing batteries which include a smaller number of higher power systems. The low exclusion probabilities of commonly used STR loci are the most negative aspect of their use, although the frequencies of both alleles of an individual can be included in calculating match. Although it is simpler and faster to perform DNA typing with STR loci than with RFLP loci and it can be performed with much smaller quantities of DNA, typing using STR loci sacrifice in exclusion power. Another disadvantage of current STR multiplex DNA typing systems is that the amplification is rarely, if ever, clean. In

other words there is considerable formation of spurious bands, which is thought to be due to DNA polymerase slippage and mis-priming events (see e.g., Tautz D., Hyper variability of Simple Sequences as a General Source for Polymorphic DNA Markers, Nuc. Acids Res., 17(16) 6463-70 (1989)).

[0011] Other polymorphisms take the form of single nucleotide variations between individuals of the same species. Such polymorphisms are far more frequent than RFLPS, STRs and VNTRs. Some single nucleotide polymorphisms occur in protein-coding sequences, in which case, one of the polymorphic forms may give rise to the expression of a defective or other variant protein and, potentially, a genetic disease. Other single nucleotide polymorphisms occur in noncoding regions. Some of these polymorphisms may also result in defective protein expression (e.g., as a result of defective splicing). Other single nucleotide polymorphisms have no phenotypic effects.

Single nucleotide polymorphisms (SNPs) can be used in the same manner as RFLPs, and VNTRs but offer several advantages. Single nucleotide polymorphisms occur with greater frequency and are spaced more uniformly throughout the genome than other forms of polymorphism. The greater frequency and uniformity of single nucleotide polymorphisms means that there is a greater probability that such a polymorphism will be found in close proximity to a genetic locus of interest than would be the case for other polymorphisms. Also, the different forms of characterized single nucleotide polymorphisms are often easier to distinguish than other types of polymorphism, *e.g.*, by use of assays employing allele-specific hybridization probes or primers).

[0013] There is a need in the art for a very accurate genetic relationship test procedure which uses very small amounts of an original DNA sample, yet produces very accurate results. This is particularly true in the forensic medicine area and criminology because often only very small samples of DNA available.

SUMMARY OF THE INVENTION

[0014] The present invention provides novel polymorphisms on the Y chromosome and methods of using Y chromosome polymorphisms as indicators of evolutionary heritage. The polymorphisms of particular interest in the present invention are clustered to specific regions of the Y chromosome, with polymorphisms of particular use found mostly in the Non-recombining Region of the human Y chromosome (NRY). These polymorphisms, including but not limited to SNPs, insertions, and deletions, may be useful for numerous applications, including forensics, paternity testing, diagnosis and the like.

[0015] In one embodiment, the present invention provides nucleic acid segments of between 10 and 100 bases containing at least 10, 15 or 20 contiguous nucleotides from any of the polymorphic regions of the Y chromosome shown in TABLE 1, and may include a polymorphic site. Complements of these segments are also included. The segments can be DNA or RNA, and can be double or single-stranded. Some segments are 10-20 or 10-50 bases long and may be less than 20 or 50 bases long. Preferred nucleic acid segments allow for the identification and analysis of nucleic acid sequences on the Y chromosome which include at least one polymorphic site that is at least diallelic.

[0016] The invention further provides allele-specific oligonucleotides that hybridize to a polymorphic region marker (M1 to M319 (excluding unassigned markers) of the Y chromosome as shown in TABLE 1, or its complement. These oligonucleotides can be probes or primers. In a particular embodiment, the nucleic acid segments include the forward and/or reverse primer sequences (e.g. primer pairs) as in Table 1. Primer pairs allow for the amplification and identification of specific polymorphic regions of the Y chromosome. Polymorphic regions of interest for amplification and/or identification include but are not limited to the NRY regions of the Y chromosome. The polymorphic regions (polymorphic markers) shown in TABLE 1 are nucleic acids of about between 100 and 700 bases, about 200 to about 600 bases and, in some embodiments, about 250 to about 500 bases in length. Many of the polymorphic nucleic acids (polymorphic

regions (markers) shown in TABLE 1 may include more than one polypmorphic site.

The invention further provides a method of analyzing a nucleic acid from an individual. The method determines which base is present at any one of the polymorphic sites of the Y chromosome as shown in TABLE 1 in bold type. Optionally, a set of bases occupying a set of the polymorphic sites shown in TABLE 1 is determined. This type of analysis can be performed on a plurality of individuals who are tested for the presence of a particular polymorphism by identifying specific polymorphic markers. The polymorphism can be correlated with a base or set of bases present at the polymorphic sites in the individuals tested, and the evolutionary heritage of the individual can be indicated by the presence or absence of a particular polymorphism.

In one embodiment, the invention provides a method for determining the ethnic origin of a male, comprising obtaining a nucleic acid sample from the male and identifying at least two polymorphic markers in the nucleic acid sample indicative of the ethnic origin of the male, using at least one primer pair from TABLE 1. The identifying of the polymorphic markers may indicate the ethnic origin of the male as being at least one of the haplotype groups selected from the group consisting of haplotype Group I, Group II, Group III, Group IV, Group V, Group VI, Group VIII, Group VIII, Group IX or Group X. In some embodiments, at least one polymorphic marker identified is a polymorphic marker from TABLE 1. The polymorphic markers may identify a haplotype associated with a haplotype group selected from the group consisting of haplotype Group I, Group II, Group III, Group IV, Group V, Group VI, Group VIII, Group VIII, Group IX or Group X, or a sub-haplotype group for the ethnic origin of the male.

[0019] In another embodiment, the invention provides a method for identifying a plurality of polymorphic sites in a nucleic acid, comprising obtaining a sample of the nucleic acid from at least one individual, and identifying, in the nucleic acid, at least one of the polymorphic sites in at least two polymorphic markers of

TABLE 1. The sample of nucleic acids may be obtained from a plurality of individuals, with the presence of the polymorphic markers in each sample of the nucleic acid determined for each of the individuals. The method may further comprise testing each individual for presence of a group of polymorphic markers which identify the haplotype of each individual, wherein the haplotype is indicative of a geographic distribution of a population or an ancestral population.

[0020] In still other embodiments, the invention provides a method for determining the ethnic origin of a human male individual, comprising obtaining a nucleic acid sample from the male, testing the nucleic acid sample for presence of a plurality of polymorphic markers selected from TABLE 1, identifying which polymorphic markers are present in the nucleic acid sample, and assigning a haplotype group to the male based on the identified markers, wherein the haplotype group is indicative of the ethnic origin of the male.

[0021] In certain embodiments, the invention provides a method for determining the paternity of a human male individual, comprising obtaining a nucleic acid sample from the male, testing the nucleic acid sample for the presence of a plurality of polymorphic markers from TABLE 1, identifying which polymorphic markers are present in the nucleic acid sample, and comparing the identified polymorphic markers to a set of polymorphic markers identified in nucleic acid samples from potential fathers.

[0022] The invention additionally provides a kit for determining ethnic origin of an individual, comprising at least two primer pairs capable of identifying at least two polymorphic markers from TABLE 1. The kit may further comprise a control nucleic acid for detecting the presence or absence of the polymorphic markers from TABLE 1.

[0023] The invention further comprises a set of primers and enzymes useful in performing an assay to identify particular polymorphisms in human male DNA.

A method of identifying polymorphisms is disclosed whereby a sample is provided and subjected to amplification using primers of the invention and thereafter determining sequences (polymorphic regions) which were amplified.

- [0024] A feature of the invention is that polymorphisms not previously identified are described herein, and are associated with a particular haplotype, indicative of a specific evolutionary heritage.
- [0025] An advantage of the invention is that the sequences disclosed herein can be used in a range of different assay systems to determine the presence of a polymorphism in a sample.
- [0026] A feature of the invention is a method for analyzing a set of unique polymorphisms on the Y chromosome to determine and identify an individual's evolutionary heritage and/or ethnicity.
- [0027] A feature of the invention is to provide a kit for determining an individual's geographical or ethnic origins.
- [0028] These and other objects, advantages, and features of the invention will become apparent to those persons skilled in the art upon reading the details of the invention as fully described below.

BRIEF DESCRIPTION OF THE DRAWINGS

- [0029] Fig. 1. Contemporary worldwide distribution of Y chromosome groups in 22 regions determined by the methods and compositions of the invention.
- [0030] Fig. 2. A phylogenetic tree deduced from 167 NRY polymorphisms on the principle of maximum parsimony.
- [0031] Fig. 3. Maximum likelihood network inferred from the haplotype frequencies.
- [0032] Fig. 4. Maximum parsimony phylogeny of human NRY chromosome biallelic variation.

DETAILED DESCRIPTION OF PREFERRED EMBODIMENTS

[0033] Before the present polymorphisms and detection methods are described, it is to be understood that this invention is not limited to particular methods or polymorphisms described, as such may, of course, vary. It is also to be understood that the terminology used herein is for the purpose of describing particular embodiments only, and is not intended to be limiting, since the scope of the present invention will be limited only by the appended claims.

Where a range of values is provided, it is understood that each intervening value, to the tenth of the unit of the lower limit unless the context clearly dictates otherwise, between the upper and lower limits of that range is also specifically disclosed. Each smaller range between any stated value or intervening value in a stated range and any other stated or intervening value in that stated range is encompassed within the invention. The upper and lower limits of these smaller ranges may independently be included or excluded in the range, and each range where either, neither or both limits are included in the smaller ranges is also encompassed within the invention, subject to any specifically excluded limit in the stated range. Where the stated range includes one or both of the limits, ranges excluding either or both of those included limits are also included in the invention.

[0035] Unless defined otherwise, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention belongs. Although any methods and materials similar or equivalent to those described herein can be used in the practice or testing of the present invention, the preferred methods and materials are now described. All publications mentioned herein are incorporated herein by reference to disclose and describe the methods and/or materials in connection with which the publications are cited.

[0036] It must be noted that as used herein and in the appended claims, the singular forms "a", "and", and "the" include plural referents unless the context

clearly dictates otherwise. Thus, for example, reference to "a nucleic acid" includes a plurality of such nucleic acids and reference to "the primer" includes reference to one or more primers and equivalents thereof known to those skilled in the art, and so forth.

[0037] The publications discussed herein are provided solely for their disclosure prior to the filing date of the present application. Nothing herein is to be construed as an admission that the present invention is not entitled to antedate such publication by virtue of prior invention. Further, the dates of publication provided may be different from the actual publication dates which may need to be independently confirmed.

THE INVENTION IN GENERAL

[0038]The use of certain nucleotide repeat polymorphisms for identifying or comparing DNA segments have been described. (See e.g., Weber & May Am Hum Genet 44:388 (1989), Litt & Luthy Am Hum Genet 44:397(1989)). The present invention is based on the finding that particular polymorphisms on the Y chromosome, including the novel polymorphisms included herein, are indicative of the evolutionary heritage and/or a paternal lineage in an individual having a Y chromosome (e.g., a male or XXY individual). These particular polymorphic genetic segments, and primers used to identify the polymorphisms for identification and comparison purposes, correspond to regions of the Y chromosome having clustered polymorphisms that are homopolymeric in regions which exhibit a very low mutation rate. An advantage of the polymorphisms of the invention is that no recombination occurs in the regions containing these markers, and thus the accumulation of mutations is preserved as an intact This creates a genetic profile that remains intact across the haplotype. generations. If men share the same derived allele, then they are identical by descent, not just by state. While a very small amount of recurrent or revertant back mutation has been observed at some markers, these anomalies are easily recognized as such because of the high resolution of the Y tree. The recognition

of new Y-chromosome markers represents a major leap in the investigation of human genetic diversity (in male lineages, complementing the information from female lineages derived from mitochondrial DNA).

[0039] The polymorphisms and methods of the present invention provide a simple way of identifying male siblingship as well as a genetic route to identify male children by so called "genebanking" using DNA or blood, or saliva from a child. Also the Y chromosome polymorphisms can reveal patterns (estimates) of recent gene flow from one gene pool to another, i.e. admixture. The methods of the present invention make the large amount of information contained in the phylogeny of haplotypes accessible for analysis.

DEFINITIONS

[0040] The term "oligonucleotide" as used herein can be DNA, RNA, or a substituted variation of these nucleic acids. The oligonucleotide may be single- or double-stranded. Oligonucleotides can be naturally occurring or synthetic, but are typically prepared by synthetic means. Preferred oligonucleotides of the invention include segments of DNA, or their complements including any one of the polymorphic sites shown in TABLE 1. The segments are usually between 5 and 100 bases (nucleotides), and often between 5-10, 5-20, 10-20, 10-50, 20-50 or 20-100 bases. The polymorphic site can occur within any position of the segment. The segments can be from any of the allelic forms of DNA shown in TABLE 1.

[0041] The term "hybridization probes" as used herein refers to oligonucleotides capable of binding in a base-specific manner to a complementary strand of nucleic acid. Such probes include peptide nucleic acids, as described in Nielsen et al., Science 254, 1497-1500 (1991).

[0042] The term "primer" as used herein refers to an oligonucleotide having at least a single-stranded portion that is adapted to act as a point of initiation of template-directed DNA synthesis under appropriate conditions (i.e., in the presence of four different nucleoside triphosphates and an agent for polymerization, such as, DNA or RNA polymerase or reverse transcriptase) in an

appropriate buffer and at a suitable temperature. The appropriate length of a primer depends on the intended use of the primer but typically ranges from 15 to 30 nucleotides. A primer need not reflect the exact sequence of the template but must be sufficiently complementary to hybridize with a template.

[0043] The term "primer site" as used herein refers to the area of the target DNA to which a primer hybridizes. The term "primer pair" as used herein refers to a set of primers including at least one 5' upstream primer that hybridizes with the 5' end of the DNA sequence to be amplified (a forward or "for" primer) and at least one 3' downstream primer that hybridizes with the complement of the 3' end of the sequence to be amplified (a reverse or "rev" primer). Primer pairs allow for the amplification and identification of corresponding polymorphic regions.

[0044] The term "polymorphic site" is used herein to describe mutations within a nucleic acid sequence which include but are not limited to site specific mutations, insertions and deletions, these mutations being found in the nucleic acid of some individuals and not in others, e.g. the polymorphic site identifies a specific polymorphism of an individual. The present invention provides segments of nucleic acid which contain at least one polymorphic site (i.e. polymorphic region). These "polymorphic regions" of the Y chromosome can be analyzed to identify a specific polymorphic site which in turn identifies a specific polymorphism associated with certain individuals.

"polymorphic regions of the present invention are also defined as "polymorphic markers" due to their usefulness in marking (identifying specific polymorphic sites). The polymorphic markers of the present invention identify specific haplotypes in the male population, these haplotypes being indicative of a specific geographical or ethnic origin. Certain polymorphic markers which identify a polymorphism shared by a large group of individuals allow for the grouping of those haplotypes which share that marker. These more commonly found markers are found at the branch points of a phylogenetic tree and are crucial in separating individuals into unique haplotype groups. The haplotype groups have this ancestral marker which branches off from a point earlier in the

phylogenetic tree. The polymorphic markers of the present invention have identified over 171 haplotypes which can be divided into ten haplotype groups.

[0046] The term "polymorphism" as used herein refers to the occurrence of two or more genetically determined alternative sequences or alleles in a population. A polymorphic marker or site is the locus at which divergence occurs. Preferred markers have at least two alleles, each occurring at a frequency of greater than 1%, and more preferably greater than 10% or 20% of a selected population, and can be present at a frequency greater than 30% to 50% or more in selected portions of the population. A polymorphic locus may be as small as one base pair. Polymorphic markers include restriction fragment length polymorphisms, VNTR's, hypervariable regions, minisatellites, dinucleotide repeats, trinucleotide repeats, tetranucleotide repeats, simple sequence repeats, and insertion elements such as Alu. The first identified allelic form is arbitrarily designated as the reference form and other allelic forms are designated as alternative or variant alleles. Polymorphisms refer to sequence differences between a reference form and a selected allele, and encompasses single or multiple nucleotide differences which can result from nucleotide insertion(s), deletion(s), substitution(s) and/ or a combination thereof. The allelic form occurring most frequently in a selected population is sometimes referred to as the wildtype form. Diploid organisms may be homozygous or heterozygous for allelic forms. A diallelic polymorphism has two forms. A triallelic polymorphism has three forms. The term "polymorphism" as used herein refers to any detectable polymorphic site in DNA or RNA that is detectable using the present methods. The term as used herein encompasses, for example, polymorphisms associated with a disease state (i.e. mutations), "silent" polymorphisms (i.e. associated with a wild-type phenotype or in a non-coding region), and polymorphisms associated with a predisposition and/or response to treatment (i.e. a polymorphism in an allele of a gene).

[0047] The term "single nucleotide polymorphism" and "SNP" as used interchangeably herein refers to a polymorphic site occupied by a single nucleotide (i.e. single base), which is the site of variation between allelic

sequences. In general, SNPs are DNA sequence variations that occur when a single nucleotide (A, T, C or G) in the genomic sequence is altered. For example a SNP might change the DNA sequence AAGGCTAA to ATGGCTAA. SNPs can occur in both coding (gene) and noncoding regions of the genome. The site is usually preceded by and followed by highly conserved sequences of the allele (e.g., sequences that vary in less than 1/100 or 1/1000 members of the population).

[0048] A single nucleotide polymorphism usually arises due to substitution of one nucleotide for another at the polymorphic site. A transition is the replacement of one purine by another purine or one pyrimidine by another pyrimidine. A transversion is the replacement of a purine by a pyrimidine or vice versa. Single nucleotide polymorphisms can also arise from a deletion of a nucleotide or an insertion of a nucleotide relative to a reference allele. Hybridizations are usually performed under stringent conditions, for example, at a salt concentration of no more than 1M and a temperature of at least 25°C. For example, conditions of 5X SSPE (750 mM NaCl, 50 mM NaPhosphate, 5 mM EDTA, pH 7.4) and a temperature of 25°-30°C are suitable for allele-specific probe hybridizations.

[0049] The term "isolated nucleic acid" as used herein refers to a nucleic acid isolated from an individual that is the predominant species present (i.e., on a molar basis it is more abundant than any other individual species in the composition). Preferably, an isolated nucleic acid comprises at least about 50, 80 or 90 percent (on a molar basis) of all macromolecular species present. Most preferably, the object species is purified to essential homogeneity, i.e. contaminant species cannot be detected in the composition by conventional detection methods. The isolated nucleic acid includes a selected DNA fragment (e.g., isolated by an amplification reaction), and an isolated mRNA.

[0050] The term "evolutionary heritage" as used herein refers to the association of a particular polymorphism with a population having a particular geographic distribution. This includes polymorphisms that are indicative of an ancestral population, i.e. a population from which an individual is a descendant.

GENERAL ASPECTS OF THE INVENTION

[0051] The present application provides novel polymorphisms, including polymorphisms clustered in and around a non-recombining portion of the human Y chromosome (NRY) The polymorphic sites and the regions flanking these polymorphic sites are shown in TABLE 1.

[0052] By knowing sequences which include particular polymorphisms on the Y chromosome, primers based on these sequences can be used in detection assays. The primers can be provided in assay kits which cover from one to any and all of the polymorphisms developed here and the kits may further comprise appropriate enzymes for use with the primers and/or reagents for the isolation and processing of nucleic acids from an individual.

[0053] The methods and compositions of the present invention allow for the genetic typing of male individuals into ten major haplotype groups. The markers and primer sets shown in TABLE 1 allow not only for typing males into one of the haplotype groups or a combination of haplotype groups, but also enables an individual to be identified to a specific geographical area associated with haplotype group. Figure 1 shows a contemporary worldwide frequency distribution of the 10 Y chromosome groups in 22 regions. Each group is represented by a distinguishing color. Colored sectors reflect representative group frequencies. The frequency distribution of the ten groups is based on > 1000 globally diverse samples genotyped using a hierarchical top down approach as illustrated in FIG.1 above the global map. The representative branching and frequency of polymorphic markers in TABLE 1 are also shown in FIG. 1 (individual marker numbers are not shown).

[0054] The identification of an individuals haplotype is base on identifying the presence of at least two distinct polymorphic markers (i.e. at least two distinct polymorphic sites must be identified), for example, polymorphic markers M91 and M278 identify haplotype 9(shown in FIG. 2 and FIG. 4). More likely, determining the haplotype of an individual involves the identification of 3 or more

markers, usually at least about 3 to 7 markers, or 7 to 9 markers or even 9 or more markers.

[0055] Haplotype groups comprise haplotypes which have at least one ancestral marker which branches off from a point earlier in the phylogenetic tree. For example, marker 91 (M91) identifies haplotypes in Group I while haplotypes in group V are identified by one marker from each of the following sets of markers; one marker from {M42, M94, M139, M251, M299} plus one from {M168, M294} and one marker from {RPS4Y, M216, M316}. To determine which haplotype group and individual is associated with, the individuals nucleic acid would need to be analyzed with at least eleven polymorphic markers. For exemplary purposes, an individuals nucleic acid could be assayed for the presence and absence of the following markers; M91, M299, M249, M294, M203, M96, M316, M9, M74, M207, M214 to determine which haplotype group they are associated with which is indicative of a certain geographical or ethnic origin.

Fig. 1 illustrates that haplotype Group I is mainly associated with Africa and in particular, southern and eastern Africa (approximately about 90% of males of haplotype Group I are of African origin). Haplotype Groups II (about 80% to about 99% frequency distribution (f.d.)) and III (about 75% to about 95% f.d.) are also strongly related to Africa compared to Groups IV through X. Populations represented in Groups I and II include some Khoisan and Bantu speakers from South Africa, Pygmies from central Africa, and lineages in Sudan, Ethiopia and Mali. Virtually all men with Group I and II haplotypes are of African affiliation from a paternal perspective. Group III lineages are predominantly African, although a sub-set of Group III lineages occur in populations bordering the Mediterranean (Middle East, Turkey, North Africa, Southern Europe).

[0057] Approximately about 70% to about 99% of the males in Group IV are of Japanese origin. Group V is slightly associated with Japan (about 10% to about 25% f.d.) and Indonesia (about 10% to about 35% frequency) with the largest frequency being associated with Australia and central Asians (about 45% to about 75% f.d.).

[0058] Group VI is more widely distributed than other haplotypes, covering the geographical area of Europe, Eastern Europe, Asia, and India. The presence of haplotype group VI in North America, Australia and Polynesia is a consequence of recent human movements since C. Columbus catalyzed the age of exploration. The largest Group VI frequency is associated with southern Europe and the middle east, with a distribution frequency of about 60% to about 85%.

Group VII is more widely associated with eastern Asia and Indonesia with distribution frequencies ranging from about 75% to about 99%. Group VIII is almost exclusively found in Papua-New Guniea (distribution frequencies of about 70% to about 95%) with a slight distribution in central Asia (distribution frequency of about 1% to about 30%). Recently, there is evidence which indicates the presence of Group VIII in Indonesia. Other specific Group VIII lineages occur in India and Europe. Individuals of haplotype Group IX are mostly associated Europe (about 75% to about 95% f.d.), India (about 25% to about 50% f.d.). Their occurrence in North America (about 35% to about 55%) Australia (35%), Polynesia is a consequence of European gene flow during the last 500 years.

[0060] Group X individuals are geographically associated with Central Asia and the Americas with a frequency distribution in North America of about 25% to about 50%, Central America of about 75% to about 95% and in South America of about 80% to about 99%. The above distribution frequencies of the various haplotypes in the geographic regions mentioned above are only representative ranges of the haplotype frequencies worldwide.

Analysis of Polymorphisms

[0061] Polymorphisms are detected in a target nucleic acid from an individual being analyzed. For assay of genomic DNA, virtually any biological sample (other than pure red blood cells) is suitable. For example, convenient tissue samples include whole blood, semen, saliva, tears, urine, fecal material, sweat, buccal, skin and hair. For assay of cDNA or mRNA, the tissue sample must be obtained from an organ in which the target nucleic acid is expressed. For

purposes of the present invention, the sample is obtained from a male, and preferably a human male.

Many of the methods described below require amplification of DNA from target samples. This can be accomplished by *e.g.*, PCR. See generally PCR Technology: Principles and Applications for DNA Amplification (ed. H. A. Erlich, Freeman Press, N.Y., N.Y., 1992); PCR Protocols: A Guide to Methods and Applications (eds. Innis, et al., Academic Press, San Diego, Calif., 1990); Mattila et al., Nucleic Acids Res. 19, 4967 (1991); Eckert et al., PCR Methods and Applications 1, 17 (1991); PCR (eds. McPherson et al., IRL Press, Oxford); and U.S. Pat. No. 4,683,202.

[0063] Other suitable amplification methods include the ligase chain reaction (LCR) (see Wu and Wallace, Genomics 4, 560 (1989), Landegren et al., Science 241, 1077 (1988), transcription amplification (Kwoh et al., Proc. Natl. Acad. Sci. USA 86, 1173 (1989)), and self-sustained sequence replication (Guatelli et al., Proc. Nat. Acad. Sci. USA, 87, 1874 (1990)) and nucleic acid based sequence amplification (NASBA). The latter two amplification methods involve isothermal reactions based on isothermal transcription, which produce both single stranded RNA (ssRNA) and double stranded DNA (dsDNA) as the amplification products in a ratio of about 30 or 100 to 1, respectively.

Detection of Polymorphisms in Target DNA

[0064] There are two distinct types of analysis depending whether a polymorphism in question has already been characterized. The first type of analysis is sometimes referred to as *de novo* characterization. This analysis compares target sequences in different individuals to identify points of variation, e.g., polymorphic sites, SNPs. By analyzing groups of individuals representing the greatest ethnic diversity among humans and greatest breed and species variety in plants and animals, patterns characteristic of the most common alleles/haplotypes of the locus can be identified, and the frequencies of such populations in the population determined. Additional allelic frequencies can be determined for subpopulations characterized by criteria such as geographical

distribution and ancestral ethnicity. The *de novo* identification of the polymorphisms of the invention is described in the Examples section. The second type of analysis is determining which form(s) of a characterized polymorphism are present in individuals under test. There are a variety of suitable procedures, which are discussed in turn.

Allele-Specific Probes

[0065] The design and use of allele-specific probes for analyzing polymorphisms is described by e.g., Saiki et al., Nature 324, 163-166 (1986); Dattagupta, EP 235,726, Saiki, WO 89/11548. Allele-specific probes can be designed that hybridize to a segment of target DNA from one individual but do not hybridize to the corresponding segment from another individual due to the presence of different polymorphic forms in the respective segments from the two individuals. Hybridization conditions should be sufficiently stringent that there is a significant difference in hybridization intensity between alleles, and preferably an essentially binary response, whereby a probe hybridizes to only one of the alleles. Probes with such specificity allow for the determination of a specific base occupying a polymorphic site in a sequence of a polymorphic region. Some probes are designed to hybridize to a segment of target DNA such that the polymorphic site aligns with a central position (e.g., in a 15 mer at the 7 position; in a 16 mer, at either the 8 or 9 position) of the probe. This design of probe achieves good discrimination in hybridization between different allelic forms.

[0066] Allele-specific probes are often used in pairs, one member of a pair showing a perfect match to a reference form of a target sequence and the other member showing a perfect match to a variant form. Several pairs of probes can then be immobilized on the same support for simultaneous analysis of multiple polymorphisms within the same target sequence.

Tiling Arrays

[0067] The polymorphisms can also be identified by hybridization to nucleic acid arrays, some example of which are described by WO 95/11995. The same array

or a different array can be used for analysis of characterized polymorphisms. WO 95/11995 also describes subarrays that are optimized for detection of a variant form of a precharacterized polymorphism. Such a subarray contains probes designed to be complementary to a second reference sequence, which is an allelic variant of the first reference sequence. The second group of probes is designed by the same principles as described in the Examples except that the probes exhibit complementarily to the second reference sequence. The inclusion of a second group (or further groups) can be particular useful for analyzing short subsequences of the primary reference sequence in which multiple mutations are expected to occur within a short distance commensurate with the length of the probes (i.e., two or more mutations within 9 to 21 bases).

Allele-Specific Primers

[0068]

An allele-specific primer hybridizes to a site on target DNA overlapping a polymorphism and only primes amplification of an allelic form to which the primer exhibits perfect complementarily. See Gibbs, Nucleic Acid Res. 17, 2427-2448 (1989). This primer is used in conjunction with a second primer which hybridizes at a distal site. Amplification proceeds from the two primers leading to a detectable product signifying the particular allelic form is present. A control is usually performed with a second pair of primers, one of which shows a single base mismatch at the polymorphic site and the other of which exhibits perfect complementarily to a distal site. The single-base mismatch prevents amplification and no detectable product is formed. The method works best when the mismatch is included in the 3'-most position of the oligonucleotide aligned with the polymorphism because this position is most destabilizing to elongation from the primer. See, e.g., WO 93/22456.

Direct-Sequencing

[0069] The direct analysis of the sequence of polymorphisms of the present invention can be accomplished using either the dideoxy chain termination method or the Maxam Gilbert method (see Sambrook et al., Molecular Cloning, A

Laboratory Manual (2nd Ed., CSHP, New York 1989); Zyskind et al., Recombinant DNA Laboratory Manual, (Acad. Press, 1988)). In a preferred embodiment, the direct sequencing would be carried using fluorescent sequencing, *e.g.*, using a PE Biosystems 373A sequencer.

Denaturing Gradient Gel Electrophoresis

[0070] Amplification products generated using the polymerase chain reaction can be analyzed by the use of denaturing gradient gel electrophoresis. Different alleles can be identified based on the different sequence-dependent melting properties and electrophoretic migration of DNA in solution. Erlich, ed., PCR Technology, Principles and Applications for DNA Amplification, (W.H. Freeman and Co, New York, 1992), Chapter 7.

Single-Strand Conformation Polymorphism Analysis

[0071] Alleles of target sequences can be differentiated using single-strand conformation polymorphism analysis, which identifies base differences by alteration in electrophoretic migration of single stranded PCR products, as described in Orita et al., Proc. Nat. Acad. Sci. 86, 2766-2770 (1989). Amplified PCR products can be generated as described above, and heated or otherwise denatured, to form single stranded amplification products. Single-stranded nucleic acids may refold or form secondary structures which are partially dependent on the base sequence. The different electrophoretic mobilities of single-stranded amplification products can be related to base-sequence difference between alleles of target sequences.

<u>Detection of SNP Polymorphisms</u>

[0072] Where the polymorphism is a SNP, any suitable method known in the art can be used in their detection. For example, the present methods can utilize the detection of SNPs by DHPLC (see U.S. Pat. No. 5,795,976) to isolate and analyze specific SNPs on the Y chromosome of a large number of individuals in a fast, efficient and inexpensive manner. This method involves separating heteroduplex

and homoduplex nucleic acid molecules (e.g., DNA or RNA) in a mixture using high performance liquid chromatography under partially denaturing conditions. In a preferred embodiment, the SNPs are identified on the Y chromosome using techniques such as those disclosed in co-pending application US Application Serial No. 09/502,558, February 10, 2000.

Mass Spectrometry

[0073] Mass spectrometry can also be used in the methods of the present invention to verify a polymorphism and/or to identify additional polymorphisms. The mass spectrum of a nucleic acid containing the polymorphic site can be compared to the mass spectrum of nucleic acids obtained from samples of known residues at the polymorphic site. These known spectra are referred to as "signature" spectra. A simple comparison of the sample spectrum vs. signature spectra will reveal whether an individual's DNA has a specific base occupying the polymorphic site. Although sequencing of fragments of nucleic acids is possible using mass spectrometry, actual sequencing of the nucleic acid is not required for this mutational analysis. Less preparation and analysis is needed to prepare and analyze a complete, intact fragment as compared to treating a sample for actual sequencing.

[0074] Certain mass spectrometry techniques can be used to analyze for polymorphisms. Short oligomers, *e.g.*, from one nucleotide up to approximately 50 nucleotides, can be analyzed and the resulting spectra compared with signature spectra of samples known to be wild-type or to contain a known polymorphism. A comparison of the locations (mass) and heights (relative amounts) of peaks in the sample with the known signature spectra indicate what type of polymorphism, if any, is present. Exemplary protocols are described in U.S. Pat Nos. 5,872,003, 5,869,242, 5,851,765, 5,622,824, and 5,605, 798, which are incorporated herein by reference for teaching such techniques.

[0075] After determining polymorphic form(s) present in an individual at one or more polymorphic site on the Y chromosome, this information can be used in a number of methods.

Methods of Use of the Polymorphisms of the Invention

[0076] The methods of the invention have utility in a wide variety of fields where it is desirable to identify known polymorphisms of a particular individual and/or to determine allelic distribution in a group or population. Such methods include, but are not limited to, linkage analysis for the identification of disease loci, evolutionary studies to determine rates of evolution in a population, identification of polymorphisms useful in forensic identification, identification of mutations associated with a disease or predisposition, genetic marker development, and the like.

Forensics

[0077] Determination of which polymorphic sites an individual possesses, identifies a haplotype, which refers to a set of polymorphic markers that distinguishes the individual. See generally National Research Council, The Evaluation of Forensic DNA Evidence (Eds. Pollard et al., National Academy Press, DC, 1996). Since the polymorphic sites of the invention are generally within a region of about 50,000 bp in the human genome, the probability of recombination between these polymorphic sites is low. The more sites that are analyzed the lower the probability that the set of polymorphic markers for one individual is the same as that in an unrelated individual. If multiple polymorphic sites are analyzed, the sites are usually in different polymorphic regions (on different polymorphic markers). Thus, polymorphisms of the invention may be used in conjunction with polymorphisms in distal genes. Preferred polymorphisms for use in forensics are diallelic because the population frequencies of two polymorphic forms can usually be determined with greater accuracy than those of multiple polymorphic forms at multi-allelic loci.

[0078] An exemplary set of polymorphic markers useful for identifying the haplotype group of an individual are the following; Markers 304(Group VI, Mediterranean), 242 (Group X, C. Asia, India, Americas), 269 (Group IX, W. Europe), 207 (Group IX, Europe, W. Asia), 74 (Groups IX-X, global), 214 (Group VII, E. Asia), 9 (Groups VII-X, global), 235 (Groups VI-X, global), 316 (Group V, Asia, America, Polynesia, Melanesia), 174 (Group IV, Asia, Japan), 299 (Groups II-X, global), 246 (Group I, Africa), 249 (Group II, Africa) 294 (Groups III-X, global), 96 (Group III, Africa, Mediterranean).

[0079] The capacity to identify a distinguishing or unique set of forensic markers in an individual is useful for forensic analysis. For example, one can determine whether a blood sample from a suspect matches a blood or other tissue sample from a crime scene by determining whether the set of polymorphic forms occupying selected polymorphic sites is the same in the suspect and the sample. If the set of polymorphic markers does not match between a suspect and a sample, it can be concluded (barring experimental error) that the suspect was not the source of the sample. If the set of markers does match, one can conclude that the DNA from the suspect is consistent with that found at the crime scene. If frequencies of the polymorphic forms at the loci tested have been determined (e.g., by analysis of a suitable population of individuals), one can perform a statistical analysis to determine the probability that a match of suspect and crime scene sample would If several polymorphic loci are tested, the cumulative occur by chance. probability of non-identity for random individuals becomes very high (e.g., one billion to one). Such probabilities can be taken into account together with other evidence in determining the innocence or guilt of an individual suspected of a criminal act.

[0080] The polymorphisms of the present invention are especially useful in identifying samples having genetic material from multiple individuals, since the polymorphisms are single copy. Thus, the detection of more than one polymorphic Y chromosome allele in a single sample is indicative of the presence of nucleic acids from multiple individuals within the sample. Such information can be useful, for example, when multiple perpetrators are suspected of

participating in a crime, or in the case of mixed unidentified remains at a grave site or accident scene.

[0081] The polymorphic sites and methods of the present invention are also useful in categorizing victims of violent crimes into ethnic and geographical groups. When a large number of victims need to be identified at a crime site, categorizing recovered victims by ethnicity can decrease the overall time for victim identification by reducing the number of comparison samples (samples from members of the victims family) to those of similar geographical origin.

Paternity Testing

[0082] The object of paternity testing is usually to determine whether a male is the father of a child. In most cases, the mother of the child is known and thus, the mother's contribution to the child's genotype can be traced. Paternity testing investigates whether the part of the child's genotype not attributable to the mother is consistent with that of the putative father. Paternity testing can be performed by analyzing sets of polymorphisms (polymorphic markers) in the putative father and the child. The polymorphic markers of the present invention can be useful in determining paternity of a male child, as they are specific to the Y chromosome. The mother need not be tested in such a case, as the mother has no contribution to the child's genotype as it pertains to the Y chromosome.

[0083] If the set of polymorphisms in the child attributable to the father does not match the putative father, it can be concluded, barring experimental error, that the putative father is not the real father. If the set of polymorphisms in the child attributable to the father does match the set of polymorphisms of the putative father, a statistical calculation can be performed to determine the probability of coincidental match. An exemplary method of determining the probability of parentage exclusion, i.e. the probability that a random male will have a polymorphic form at a given polymorphic site that makes him incompatible as the father) is described in WO 95/12607.

[0084] If several polymorphic loci are included in the analysis, the cumulative probability of exclusion of a random male is very high. This probability can be

taken into account in assessing the liability of a putative father whose polymorphic marker set matches the child's polymorphic marker set attributable to his father. This analysis can be further expanded to identify ancestral males (e.g., grandfather, great grandfather and so on). Such analysis can be useful in genealogical analysis, or in tracing the origin of ancestral man (e.g.) using samples obtained from an archeological site).

Longer-term Family Heritage

In addition to the use in paternity testing, the polymorphisms and methods of the present invention can be used to determine relationships through a paternal lineage for multiple generations. The constancy and low mutational rate of these regions of the Y chromosome allow an individual to trace his specific ancestral lineage using the Y chromosome polymorphisms. For example, a specific residue (base) in a polymorphic site may be indicative of a population that is in or from a certain region in Europe. Assaying an individual for this polymorphism can indicate that the individual's paternal ancestors were in or descended from this particular region.

Correlation of Polymorphisms with Phenotypic Traits

[0086] The polymorphisms of the invention may contribute to the phenotype of an organism in different ways. Some polymorphisms occur within a protein coding sequence and contribute to phenotype by affecting protein structure. The effect may be neutral, beneficial or detrimental, or both beneficial and detrimental, depending on the circumstances. Other polymorphisms occur in noncoding regions but may exert phenotypic effects indirectly via influence on replication, transcription, and translation.

[0087] A single polymorphism may affect more than one phenotypic trait. Likewise, a single phenotypic trait may be affected by polymorphisms in different genes. Further, some polymorphisms predispose an individual to a distinct mutation that is causally related to a certain phenotype.

[0088] Phenotypic traits include diseases that have known but hitherto unmapped genetic components. Phenotypic traits also include symptoms of, or susceptibility to, multifactorial diseases of which a component is or may be genetic, such as autoimmune diseases, inflammation, cancer, diseases of the nervous system, and infection by pathogenic microorganisms. Phenotypic traits also include characteristics such as longevity, appearance (e.g., baldness, obesity), strength, speed, endurance, fertility, and susceptibility or receptivity to particular drugs or therapeutic treatments.

[0089] Correlation is performed for a population of individuals who have been tested for the presence or absence of a phenotypic trait of interest and for polymorphic markers sets. To perform such analysis, the presence or absence of a set of polymorphisms (i.e. a polymorphic set) is determined for a set of the individuals, some of whom exhibit a particular trait, and some of which exhibit lack of the trait. The alleles of each polymorphism of the set are then reviewed to determine whether the presence or absence of a particular allele is associated with the trait of interest. Correlation can be performed by standard statistical methods such as a κ-squared test and statistically significant correlations between polymorphic form(s) and phenotypic characteristics are noted.

[0090] The polymorphisms and assays of the present invention are of particular use in determining the appropriate populations for mapping complex genetic traits and/or disorders. Population choice can be crucial for the success of gene mapping for particular traits and/or disorders. Populations having a high degree of inbreeding are also useful for linkage analysis (see, *e.g.*, Sheffield, VC et al., *Trends in Genetics* 4:391-6 (1998)), and the polymorphisms of the invention can be useful in determining the genetic heterogeneity of a population.

Antibodies to Specific Polymorphisms

[0091] Polyclonal and/or monoclonal antibodies that specifically bind to variant gene products but not to corresponding prototypical gene products are also provided. Antibodies can be made by injecting mice or other animals with the variant gene product or synthetic peptide fragments thereof. Monoclonal

antibodies are screened as are described, for example, in Harlow & Lane, Antibodies, A Laboratory Manual, Cold Spring Harbor Press, New York (1988); Goding, Monoclonal antibodies, Principles and Practice (2d ed.) Academic Press, New York (1986). Monoclonal antibodies are tested for specific immunoreactivity with a variant gene product and lack of immunoreactivity to the corresponding prototypical gene product. These antibodies are useful in diagnostic assays for detection of the variant form, or as an active ingredient in a pharmaceutical composition.

Use of the Present Method to Produce a Database of Y Chromosome Polymorphisms

[0092] The polymorphisms of the invention can be used as the basis for, or combined with other such polymorphisms to provide, a general catalog of genome variation to address the large-scale sampling designs required by association studies, gene mapping, and evolutionary biology. There is widespread interest in documenting the amount and geographic distribution of genetic variation in the human species. This information is desired by the biomedical community, whose work would be greatly facilitated by a densely packed map of polymorphic markers, particularly SNPs in the NRY region, to be used to for example, identify genes associated with disease by linkage disequilibrium between sets of adjacent markers and the occurrence of disease in populations, and to characterize disease-related variation among populations.

[0093] Anthropologists and archeologists use genetic variation to reconstruct our species' history, and to understand the role of culture and geography in the global distribution of human variation. The requirements for these two perspectives seem to be converging on a need for an accessible, representative DNA bank and statistical database of human variation.

[0094] In addition, these systems have potential in both routine forensic and intelligence database applications, either in place of or in conjunction with more traditional "DNA fingerprinting" databases produced using methods such as restriction fragment length polymorphism mapping.

[0095] The invention may be embodied in computer-readable media containing an electronically, magnetically, or optically stored code representative of the markers for polymorphic regions of Table 1, and/or stored code configured to create the electronically stored representation of Table 1 and the corresponding geographic distributions for these polymorphic markers (see TABLE 3). Such databases may be produced using a variety of different data configurations and processing capabilities. Examples include, but are not limited to, logical databases, physical databases, relational databases, central configuration databases, and the like. Database structures for genomic information may be based on, for example, the database structures disclosed in U.S. Patent No. 6,229,911. In other examples, the data generated for use in the present invention may be used to create a general database such as that described in U.S. Pat. No. 4,970,672 or a relational database such as that described in U.S. Pat. No. 5,884,311. Databases containing data generated for use in the methods of the invention may also be a central configuration database for data that is shared among multiprocessor computer systems. See U.S. Pat. No. 6,014,669. Other database systems and design methodologies can be found in I. Fogg and M. Orlowska, Computers Math. Applic. (UK), (1993) 25:97-106; S. Ceri, et al., Proceedings of the IEEE (1987) 75:533-545.

EXAMPLES

Skill in the art with a complete disclosure and description of how to make and use the present invention, and are not intended to limit the scope of what the inventors regard as their invention nor are they intended to represent that the experiments below are all or the only experiments performed. Efforts have been made to ensure accuracy with respect to numbers used (e.g. amounts, temperature, etc.) but some experimental errors and deviations should be accounted for. Unless indicated otherwise, parts are parts by weight, molecular weight is weight average molecular weight, temperature is in degrees Centigrade, and pressure is at or near atmospheric.

EXAMPLE 1

[0097] A phylogenetic tree was deduced from 167 polymorphisms from a Nonrecombining Region of the human Y chromosome (NRY)on the principle of maximum parsimony (Figure 2). Seven of the 167 polymorphisms had been detected by means other than DHPLC and were taken from the literature to demonstrate the applicability of the method of the invention to polymorphisms with less demographic specificity than those in TABLE 1. Seventy-three of the 160 polymorphisms detected by DHPLC had been reported previously. Underhill, P. A. et al Genome Res. 7:996-1005 (1997). Shen, P. et al Proc. Natl. Acad. Sci. USA 97:7354-7359 (2000). Of the remaining 87 unreported polymorphisms, 53 were discovered in a set of 53 individuals of diverse geographic origin during the screening of the unique sequences and repeat elements, other than long interspersed elements, contained in three overlapping cosmid sequences (GenBank accession nos. AC003032, AC003095, AC003097) and a few small fragments scattered throughout the NRY. Finally, 34 were detected during genotyping. In total, the marker panel comprises 91 transitions, 53 transversions, 22 small insertions or deletions, and an Alu insertion. All polymorphisms are biallelic, except a double transversion, M116, that has three alleles, A, C or T, defining quite different haplotypes. Two non-CpG associated transitions (M64 and M108) showed evidence of recurrence but generated no ambiguities when considered in the context of other markers. The primer sequences used to detect the 167 polymorphisms are given in Table 1).

METHODS

samples with their subsequently determined haplogroup designations: *Africa*: 3 Central African Republic Biaka II, III (1); 2 Zaire Mbuti II, III; 2 Lissongo II, III; 2 Khoisan I, III; 1 Berta VI; 1 Surma I; 1 Mali Tuareg III; 1 Mali Bozo III; *Europe*: | Sardinian VI; 2 Italian VI IX; 1 German VI; 3 Basque VI, IX (2); *Asia*: 3 Japanese IV, V, VII; 2 Han Chinese VII, 1 Taiwan Atayal VII, 1 Taiwan Ami,

VII, 2 Cambodian VI, VII; *Pakistan*: 2 Hunza VI, IX; 2 Pathan VI, VII; 1 Brahui VIII; 1 Baloochi VI; 3 Sindhi III, VI, VIII; *Central Asia* 2 Arab IX; 1 Uzbek IX; 1 Kazak V; *MidEast*: 1 Druze VI; *Pacific*: 2 New Guinean V, VIII; 2 Bougainville Islanders VIII; 2 Australian VI, X: America: 1 Brazil Surui, 1 Brazil Karatina, 1 Columbian, 1 Mayan all X. An additional 1,009 chromosomes, representing 21 geographic regions, were genotyped by DHPLC for all markers other than those on the terminal branches of the phylogeny. The latter were genotyped only in individuals from the haplogroup to which those markers belonged. This hierarchic genotyping protocol was necessitated by the minute amounts of genomic DNA available for most samples.

[0099] PCR. The RepeatMasker2 program (http://ftp.genome.washington.edu) was used to identify human repeat DNA sequences. Primers were designed to amplify unique sequences and repeat elements other than LINE as confirmed by a negative female control, yielding amplicons 300-500 bp in length. All primers had a uniform annealing temperature, which allowed a single PCR protocol to be used. It comprised an initial denaturation at 95°C for 10 min to activate AmpliTaq Gold®, 14 cycles of denaturation at 94°C for 20s, primer annealing at 63-56°C using 0.5°C decrements, and extension at 72°C for 1 min, followed by 20 cycles at 94°C for 20 s, 56°C for 1 min, and 72°C for 1 min, and a final 5-min extension at 72°C. Each 50-μl PCR reaction contained 1 U of AmpliTaq Gold® polymerase, 10 mM Tris-HCl, pH 8.3, 50 mM KCl, 2.5 mM MgCl₂, 0.1 mM each of the four deoxyribonucleotide triphosphates, 0.2 μM each of forward/reverse primers, and 50 ng of genomic DNA. PCR yields were determined semi-quantitatively on ethidium bromide stained agarose gels.

[00100] DHPLC analysis. Unpurified PCR products were mixed at an equimolar ratio with a reference Y chromosome and subjected to a 3-minute 95°C denaturing step followed by gradual reannealing from 95 to 65°C over 30 min. Ten microliters of each mixture were loaded onto a DNASepTM column (Transgenomic, San Jose, CA), and the amplicons were eluted in 0.1 M triethylammonium acetate, pH 7, with a linear acetonitrile gradient at a flow rate of 0.9 ml/min². Under appropriate temperature conditions, which were optimized

by computer simulation (available at http://insertion.stanford.edu/melt.html), mismatches were recognized by the appearance of two or more peaks in the elution profiles.

DNA sequencing. Polymorphic and reference PCR samples were purified with QIAGEN (Valencia, CA) QIAquick spin columns. Both strands were sequenced to determine the location and chemical nature of any polymorphic sites, using the amplimers as sequencing primers and ABI Dye-terminator cycle sequencing reagents (PE Biosystems, Foster City, CA). Each cycle sequencing reaction contained 6 μl of purified PCR product, 4 μl dye terminator reaction mix, and 0.8 μl of primer (5 μM). Cycle sequencing was started at 94°C for 1 min, followed by 25 cycles of 96°C for 10s, 50°C for 2s, and 60°C for 4 min. The sequencing products were purified with CentrifexTM gel filtration cartridges (Edge Biosystems, Gaithersburg, MD) and analyzed on a PE Biosystems 373A sequencer.

Statistical analysis. The program CONTML in PHYLIP, version 3.57c, [00102] was used to construct a frequency based maximum likelihood network. The expected Luria-Delbrück/Lea-Coulson distribution of the number of mutants for each gene was fitted by maximum likelihood, treating each nucleotide of the screened sequence as analogous to a parallel, independent bacterial culture Luria, S. E. & Delbrück, Genetics 28:491-511 (1943); Lea, D. E. & Coulson, A. C. Genetics 49:264-285 (1949). The distributions under the expectation of constant population size were calculated according to Watterson, G. A. Theor. Popul. Biol. 7: 256-276 (1975). Mismatch distributions were calculated as described previously (Shen et al., supra). The NRY mutation rate per nucleotide per year (1.53×10^{-9}) was calculated on the basis of 597 nucleotide substitution differences between human and chimpanzee observed over 39,931 bp of non-coding sequence (Shen et al., supra). The corresponding mutation rates for mtDNA (1.65 x 10⁻⁸) and X chromosome (7.54 x 10^{-10}) were calculated on the basis of 581 and 58 nucleotide substitution differences, respectively, between human and chimpanzee observed over 6,176 bp of coding mtDNA (mictochondrial DNA) sequence comprising the genes ND1, ND2, COX1, COX2, ATP8, ATP6, COX3, and ND3, and 7,853 bp of flanking non-coding sequence of the DIAPH2 gene on Xq22.

[00103] Accession numbers. Most of the NRY sequence surveyed was derived from 5 cosmid sequences retrievable from Genbank using the accession numbers AC003031, AC003032, AC003094, AC003095, and AC003097. Six polymorphisms were affiliated with genomic regions for DFFRY (AC002531), one each for DBY (AC004474) and UTY1 (AC006376), 3 for SRY (NM003140), and 15 for random genomic STSs reported by Vollrath D, et al. *Science* 258:52-59 (1992).

The tree of Figure 2 is rooted with respect to non-human primate [00104] sequences. The 116 numbered compound haplotypes were constructed from 167 mutations (markers) of which 160 were discovered by DHPLC (Table 1). Seven haplotypes from the literature with less geographical heritage specificity were also analyzed in this study, including YAP (M1), DYS271 (M2), PN3 (M29), SRY 4064 (M40), TAT (M46), RPS4YC711T (M130), and SRY 2627 (M167), (the sequences for these markers are not shown in TABLE 1). Marker numbers indicated on the segments are discontinuous because of the removal of all but one polymorrhism associated with tandem repeats and homopolymer tracts whose ancestral state is uncertain. Haplotypes are assorted into ten haplogroups (I - X) using principles commonly applied to haploid mtDNA phylogenies. Macaulay, V. et al. Am. J. Hum. Genet. 64: 232-249 (1999). Haplogroup I members, ancestral for M42, M94 and M139, also share the only homopolymer-associated marker M91. All haplogroup I individuals have an 8-T length variant, while 1,009 men in haplogroups II-X have 9 T's and in two cases 10 (not shown). Only one inconsistent haplogroup X individual had 8 T's (not shown). Haplogroups I and II, both of which are almost exclusively represented in Africa only, share the ancestral allele of M168. Haplogroup III is generally the most frequent one in Africa. Its frequency decreases with increasing distance from Africa, from 27% in the Mid-East to a few percent in Northern Europe and South and Central Asia. Haplogroup IV, related to the former through M1 and M145, is found mainly in Japan.

[00105] In a recent cladistic analysis of nine diallelic NRY polymorphisms, including M1, in 1,544 individuals, it was hypothesized that haplogroup III comprises descendents of a range expansion that brought Y-chromosomes back to Africa (M. F. Hammer et al. 15:427-441 (1998)). Haplogroups V and VIII are prevalent in New Guinea and Australia, but they are also found at varying though smaller frequencies throughout Asia. Haplogroups VI and IX are found mostly in Europe and the Indus Valley. They are not observed in East Asia, where haplogroup VII dominates, suggesting that this part of the world where agriculture developed independently resisted effectively subsequent gene flow Macaulay, V. et al. supra. The distinction between Eurasians and East Asians was also observed with mtDNA Macaulay, V. et al., supra., and autosomal genes (Diamond, J. Guns, Germs, and Steel (Norton & Co., New York, p. 99, 1999). Haplogroup X is common in the Americas, although its origin may have been in Central Asia where traces of it persist, as shown in Table 2:

TABLE 2.

Haplogroup	Exemplary Defining Mutation	Avg. no. of Mutations from Root to Individual Haplotypes	Total no. of Individuals	No. of Mutations per Haplogroup Minus Defining Mutation(s)	No. Haplotypes per Haplogroup
I	M91	6.1 ±0.95	52	20	8
II	M60	6.1±0.41	52	12	10
III	M96	10.4±0.24	218	27	21
IV	M124	10.5±0.56	9	7	4
v	M130	6.6±0.6	40	8	5
VI	M89 & absence of M9	7.4±0.25	163	25	23
VII	M175	9.5±0.35	137	18	15
VIII	M9 & Absence of M175 and M45	8.9±0.63	67	16	11
IX	M173	10.2±0.20	195	13	13
X	M74 & Absence of M173	9.2±0.1	129	6	6
Totals		8.59±0.20	1052	152	116

EXAMPLE 2

[00106] The root of the phylogeny was placed using sequence information generated from the three great ape species. The sequential succession of mutational events is unequivocal, except for those appearing in the same tree

segment (e.g., M42, M94, M139). The phylogeny is composed of 116 haplotypes and their frequencies in 21 general populations are listed in Table 3. Forty-two haplotypes (36.2%) are represented by just one individual. Several haplotypes, however, display higher frequencies and/or geographic associations that reveal patterns of population affinities apparent from a maximum likelihood analysis (Figure 3) performed on the haplotype frequencies reported in Table 3. To facilitate presentation, the 116 haplotypes were grouped into 10 haplogroups as defined either by the presence or absence of mutations occupying strategic positions in the phylogeny. Haplogroups VI, VIII, and X, although polyphyletic, are distinguished by the criteria in Table 2.

[00107] Three mutually reinforcing mutations, M42, M94 and M139 (2 transversions and a 1-bp deletion) unequivocally distinguish haplogroup I which is represented today by a minority of Africans, mainly Sudanese, Ethiopians, and Khoisans (Table 2). All non-African, except a single Sardinian, and the majority of African males sampled, carry only the derived alleles at the three sites. This implies that modern extant human Y-chromosomes trace ancestry to Africa and that the descendents of the derived lineage left Africa and eventually replaced archaic human Y-chromosomes in Eurasia.

[00108] An important property of a phylogeny is the randomness of number of mutations per segment of the tree. Forty-one of the total 166 segments carry no mutation, while 98, 16, 8, 2, and 1 segment have 1, 2, 3, 4, and 8 mutations, respectively. The mean number of mutations per segment is 1.024 with a variance of 0.945. Applying the G-test for goodness of fit and Williams' correction to the observed G, the data do not fit a Poisson distribution (G_{adj}=34.98, df=3, P~10⁻⁷). This is due to an excess of segments with one mutation, as expected in an exponentially growing population. Similar results were obtained recently for the separate analysis of 4 Y-chromosome genes. Further support that the human population has undergone a major expansion comes from the consistently negative values of Tajima's D (Lea, DE & Coulson, AC Genetics 49: 264-285 (1949)) for not only the Y-chromosome, but also for mitochondrial DNA, X-

chromosomal and autosomal genes. Interestingly, NRY shows evidence of significantly reduced variability to the other genetic systems (Shen et al., *supra*), confirming a similar comparison of a smaller number of polymorphisms on previously reported NRY sequences with eight X-linked (Hudson, R. et al, *Genetics* 116:153-159 (1987); Nachman, M. W. *Mol. Biol. Evol.* 15: 1744-1750 (1998) and 16 autosomal human genes. Possible explanations include positive selection on NRY Jaruzelska, J et al., *D. Mol. Biol. Evol.* 16:1633-1640 (1999) and a difference between male and female effective population sizes Wyckoff, G. J et al., *Nature* 403:304-309 (2000). Assuming expansion, the age of the most recent common ancestor (*T_{MRCA}*) was previously estimated at 59,000 years with a 95% probability interval of 40,000-140,000 years (Thomson, R. et al. *supra*).

[00109] This value is similar to an estimate of 46,000 to 91,000 years based on 8 Y chromosome microsatellites (Pritchard, J. K et al, *Mol. Biol. Evol.* 16:1791-1798 (1999) and, therefore, is considerably less than estimates of >100,000 years obtained previously (Hammer et al, *supra*). Of course, this assumes that selection or population structure have not had a major effect on NRY diversity, an assumption that may be wrong in light of our findings of significantly reduced variability on NRY. As the average number of mutations of all segments departing from the root is 8.60 (Table 3), and with a *T*_{MRCA} value of 59,000 years, the average time for adding a new mutation to the tree is 6,900 year. This puts the age of M168 that marks the expansion of anatomically modern humans out of Africa at approx. 44,000 years, in agreement with a previous estimate of 47,000 years with 95% probability intervals of 35,000 to 89,000 years using the program GENETREE (Thomson, R. et al. *Proc. Natl. Acad. Sci.* USA 97:7360-7365 (2000).

TABLE 3.

Haplotype Group			_						بِ	46	44		11		- 40	17	46	10 /	0 0		22	24 0	E 00	97	38	29	20 0	4 20	22	34 4	15 20	37 9	8 39
Haplotype #	1	2	3		5	6	7	8	9	10			13 1	15	16	17	15	19 2	0 2	22	23	24 2	2	21	26	1	,	1 32	. 33		7 30		2
Sudan Ethiopia	6	17	1		1					3			1							3			15				6	2			206		-
Malı	U	,			•			1	3	·	•	,	1					1	1	7					13		-					1 1	2
Morocco								•	•		•		•							2											1		1
C. Africa											1	1		1	7	1	1		1					3									
Khoisan				11		5	1								11					7											4		
S. Africa				3								7								28	1	3	2	8			1				1		
Europe																												- 1					
Sardinia		1																										1			4		
Basque																																	1
Mid-east																				2					1			1			2		1
C. Asia + Sibena																												. 2	1		1		
Pakistan + India												2															- 3	2		1			
Hunza																																	
Japan																																	
China																																	
Tawan																																	
Cambo + Laos																																	
New Guinea																																	
Australia																																	
Amenca																				2											1		
Total	6	23	1	14	- 1	5	1	1	3	3	3	19	2 1	1	18	1	1	1	1 1	71	1	3	2 17	12	14	2	19	27	1	1 :	35 11	1 1	6 1
Group		iV		Τ-		V						_						V	1									Т-			VII		
Haplotype #	41	42	43	44	45	46	47	48	49	50	51	52	53 5	4 55	56	57	58	59	50 G	62	63	64 4	5 66	67	68	69	70 7	1 72	73 7	4 75	76	7 78	79 80
Sugan																											4	ļ					
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Mali	l																																
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C. Africa																																	
Khoisan																																	
S. Africa																																	
Europe									1	1		8	1	ı			2	1									9						
Sardinia	l									11					1												2						
Basque	1									2		1															1						
Mid-east														1			2										8						
C. Asia + Sibena					10	16									2	1	12		4	1	1		2		1		1 1					6	9
Pakistan + India						1										4	3		3 2	!		1	1	1	4		7					1	1
Hunza	ļ					1											1		3				1		2		1					1	
Japan	1	5	1			1	1	1																							2	. 2	1
China																												1		1		1 2	4 1
Tawan																													4		18		_
Cambo + Laos						1																2				1			•	1		1	1
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Total	1	5	1		10	24	. 1	1	1	15	1	10	1	1	5	5	23	1	10 2	1	_1_	3	3 1	_1	7	_1_	1 6	8 1	4 .	<u> </u>	22	2 12	16 1
Group			VII							ν	111										1)									X			
Haplotype#	82	83	84	. E	86	87	88	89	90	91	92	93	94 9	5 96	97	98	98	100	101 1	02 10	3 10	4 10	5 106	107	10	3 109	110	111	112 1	13 1	14 11	5 116	Tot
Sudan	Т				-												-																40
Ethiopia																					1												88
Mak	1																				1												44
Morocco	l						1														5	i											28
C. Afnca	l																																37
Khoisan	l																																39
S. Africa	1																																53
Europe	1															3	1				29	9			3								60
Sardinia	l																	2															22
Basque																			2	7 5													45
Mid-east	1								2												2												24
C. Asia + Siberia	1	2			1	5			2	2	12	1									10			1	30	3	6	3		12	6		18
Pakistan + India	ĺ	•			•	-	1	2			_	•									6		1		28			2		4			88
Hunza	1					2	•	_	-	3											3				11			2		7			38
						-				~		1									_				- '								23
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China	5		1	I		1 1 7							2 :	, A	1										1				·				

[00110] This concurs with recent archeological and mtDNA data, and is also consistent, though at a compressed time scale, with the weak Garden of Eden hypothesis. Under this hypothesis, a small subgroup of behaviorally modern humans left Africa and separated into several fairly isolated groups represented today by the major haplogroups III-X. Those groups remained small throughout the last glaciation before they underwent roughly simultaneous expansions in size as suggested by the star-like genealogy shown in Figure 1. In conclusion, the new levels of biallelic variation revealed here suggest a recent ancestry of the paternal lineages of our species from Africa and testify to the informativeness of the Y chromosome in deciphering the evolution of humankind.

[00111] The gene frequencies of New Guineans and Australian aborigines were grouped together because of the small sample size of the latter. Values at nodes indicate number of 1,000 bootstrap trees presenting cluster distal of node. Sudanese and Ethiopians are distinct from the other Africans and appear to be more associated with samples from the Mediterranean basin. This may reflect either repeated genetic contact between Arabia and East Africa during the last 5,000 to 6,000 years or a Middle Eastern origin with subsequent acquisition of Negroid genes on the way southwest with agricultural expansion. Native Americans are located between Eurasians and East Asian indicating common ancestry with both. This network is consistent with the first two principal components capturing 18% of the variation present in the 116 haplotypes.

EXAMPLE 3

- [00112] A phylogenetic tree was deduced from NRY polymorphisms on the principle of maximum parsimony (Figure 3). Figure 3 shows the phylogenetic tree deduced from 304 polymorphisms including those presented in Examples 1 and 2 as well as other novel markers.
- [00113] The contemporary global frequency distribution of the 10 Groups based on >1000 globally diverse samples genotyped using a hierarchical top down approach is illustrated in Figure 3. 171 haplotypes are identified in Fig.3 as well as their relationship with 309. However 4 markers are recurrent but define

distinctive haplotypes when considered in the context of the other markers. The 4 markers are M64.1 (M64.2),M108.1 (M108.2), M116.1 (M116.2) and 12f2.1 (12f2.2). For example M64.1 occurs on haplotype #80 in Group V and M64.2 on ht#159 in Group IX.

[00114] The relationship of the haplotypes to the ten haplogroups is also shown in Fig. 3. Each haplotype can be related to a specific geographical region within the haplotype group, allowing for very specific geographic association and ethnic identity of male individuals. Fig. 3 also shows which specific markers are important branching points for distinguishing between haplotype groups and also sub-haplotype groups such as haplotypes 10-13 of group II. This composite collection of 315 NRY variants (polymorphic markers) provides improved resolution of extant patri-lineages.

EXAMPLE 4

[00115] The methods of the invention can be utilized in the area of forensics to determine the ethnic affiliation of an individual.

[00116] The method involves, obtaining a nucleic acid sample from the individual and processing the sample sufficiently to conduct PCR amplification on the sample. The method exploits the hierarchical property of the Y chromosome gene tree that reveals the unequivocal sequential accumulation of DNA variation during the lineal life spans of these haplotypic molecules. Since Y chromosome haplotypes display a strong correlation with geography, such data provides insights into the affinity and diversification of populations. The sample is analyzed at polymorphic sites defining key internal nodes within the phylogeny. At least 11 primers sets, with each primer set recognizing at least one polymorphic region on the Y chromosome from a different haplotype group (Group I through Group X) are required to begin localizing a sample within the phylogeny. Additional haplotype resolution can be obtained by typing a subset of related markers. Each PCR reaction carried out on the sample, may include one or more primer sets/reaction vessel.

- [00117] The PCR amplified products are analyzed by DHPLC (or any other suitable PCR product detection technique, such as DNA chips, direct sequencing, Taqman and the like) genotyping technology to define the haplotype which is then compared to a data base detailing the geographic association of the haplotype. The data base utilizes the markers identified in TABLE 1 and various combinations thereof which enables the identification of an individual to a particular haplotype group (Group 1 through Group X) as well as haplotype which are indicated in FIG.2 and FIG.4.
- [00118] In certain instances, primer sets to the following markers are utilized to identify which haplotype group an individual originates from; Markers- M91, M60, M96, M174, (M216 or M316), M89, M9, M175, M45, M173. These markers identify the following haplotype groups; Group I = M91, Group II = M60, Group III = M96, Group IV = M174, Group V = M316, Group VI = M89 without M9, Group VII = M9 without M175 or M45, Group VIII = M9, Group IX = M173 and Group X is represented by marker M74 without M173. This approach can be expanded to increase criteria for inclusion/exclusion decisions.
- [00119] TABLE 4 shows a two stage scheme of 30 markers, the haplotype groups they help define as well as geographical region associated with the haplotype group and the polymorphic markers which provides considerable power in facilitating localization any Y chromosome in the phylogeny. In cases where more than one marker is listed in TABLE 4, any one marker in the subset will provide comparable information.

TABLE 4			
Markers analyzed	Assoc. Geographical	Markers analyzed	Assoc.
Analysis #1	region	Analysis # 2	Geographical
			region
M42, M94, M251, or	Global	M215, M243, or	Africa, Med
M299 (Groups II-X)		M293 (Group III)	
M246	Africa	M2, M180 or	Sub Saharan Africa
(Group I)		M291 (Group III)	

M181 or M249	Africa	M191	Sub Saharan Africa				
(Group II)		(Group III)					
M168 or M294	Global	M35	Africa, Med,				
(Groups III-X)		(Group III)	S. Europe				
M96	Africa, Med.	M217	E. Asia, India,				
(Group III)		(Group V)	N. America,				
M174	Asia, Japan	M201	Med., S. Europe				
(Group IV)		(Group VI)					
M216 or M316	Asia, America,	M172	Med., S. Europe				
(Group V)	Polynesia, Melansia	(Group VI)					
M89, M213 or M235	Global	M267	Med., S. Europe				
(Groups VI-X)		(Group VI)					
M9	Global	M170 or M258	Europe				
(Groups VII-X)		(Group VI)					
M175 or M214	E. Asian	M52 or M69	India				
(Group VII)		(Group VI)					
M45 or M74	Global	M122	E. Asia				
(Groups IX-X)		(Group VII)					
M173 or M207	Europe, W. Asia	M119	E. Asia				
(Group IX)		(Group VII)					
M269	W. Europe	M268	E. Asia				
(Group IX)		(Group VII)					
M242	C. Asia, India,	M17 or M198	E. Europe, W. Asia				
(Group X)	Americas	(Group IX)					
M304	Med.	M3	N.& S America				
(Group VI)		(Group X)					

[00120] This example demonstrates that by using about 10% of the markers, one can localize any sample to a "neighborhood" or sub-haplotype group in the tree.

These markers are useful in identifying a male for which no ethnic origin is

known. If it was known that the individual to be typed was for example, from Peking, then the assemblage of a more "Asian" group of markers would be more useful than those in TABLE 4.

[00121] The methods of the invention allow for the ability of Y markers to define (on a general geographic or population level) male ethnic affiliation.

[00122] While the present invention has been described with reference to the specific embodiments thereof, it should be understood by those skilled in the art that various changes may be made and equivalents may be substituted without departing from the true spirit and scope of the invention. In addition, many modifications may be made to adapt a particular situation, material, composition of matter, process, process step or steps, to the objective, spirit and scope of the present invention. All such modifications are intended to be within the scope of the claims appended hereto.

TABLE 1

M2 = DYS271 (209 bp) A to G at position 168

aggcactggtcagaatgaagTGAATGGCACACAGGACAAGTCCAGACCCAGGAAGGTCCAGTAACATGGGAGAAGAACGGAAGGAGTTCTAAAATTCAGGGCTCCCTTGGGCTCCCCTGTTTAAAAATGTAGGTTTTATTATTATTATTTCATTGTTAACAAAAGTCCRTGAGATCTGTGGAGGATAAAGgggggagctgtattttccatt

For: 5'-3' = aggcactggtcagaatgaag Rev 5'-3' = aatggaaaatacagctcccc

M3 = DYS199 (241 bp) C to T at position 181

taatcagtctcctccagcaAGTGATATGCAACTGAGATTCCTTATGACACATCTGAACA CTAGTGGATTTGCTTTGTAGTAGGAACAAGGTACATTCGCGGGATAAATGTG GCCAAGTTTTATCTGCTGCCAGGGCTTTCAAATAGGTTGACCTGACAATGGGT CACCTCTGGGACTGAYAATTAGGAAGAGCTGGTACCTAAAATGAAAGATGCc cttaaatttcagattcacaatttt

For: 5'-3' = taatcagtctcctcccagca Rev 5'-3' = aaaatlgtgaatctgaaatttaagg

M4 = DYS234 (273 bp) A to G at position 88

tcctaggttatgattacagagcgAGGATTATTATAATATTGGAATAAAGAATAATTGCTACA AACTAATGATTAATGATATTCATAT**R**TAATCATATCTAAGATCTATATCTAGT ATAACTATTCTTATTTTATATATTTTATTGTACTGGAACAGCTTGTGCCCTTGG TCTCTTGCCTCGGCACCTGGGTGGCTTGCCATCCACAGAAGTGTTTTAACAGC AAAAATTACTGTGAATTTTCTGCCCAAAAccttgtcatgtttacaagacgt

For: 5'-3' = tecta.ggttatgattacagageg Rev 5'-3' = acgtettgtaaacatgacaagg

M5 = DYS214a (322 bp) C to T at position 73

gggtttatactgacctgccaatgttAAAAGGGACCTAAATTCACTTTGGGGAAGTGGCCAGA AAGGAAGAAGYAGAAGGAGAAGAGTGCAAGAAACCTCCAGTTGTGGGGGTT GAGCCTCCACGATAAGAAAGAAAGAAATCTCCAGTAGGGGGGATTGAGCCT AACACAAACCTTTGGTAATAGACAAGGCAAGACATTTCCAATAGGGGAGATT GAGTGTCACCTCAAAACTATTAAGATGGGAAATACCCCAGGTAAGATAGAGG GTAAAAAAGGATAAAGCTAGCAGCAATAACATTCcccctgaaagttcccaataa

For: 5'-3' = gggtttatactgacctgccaatgtt

Rev 5'-3' = ttattgggaactttcagggg

DYS214 complete. (656 bp) This fragment was converted into two STSs, a & b, containing M4 and M16 respectively. The two new STSs (a & b) omit an extra internal 68 bp region within the complete STS.

For 3'-5' = gggttlatactgacctgccaatgtt

Rev 5'-3' = ccgtgtgttgctgggctgtc

M6 = DYS198 (2.18 bp) **T** to **C** at position 37

CactaccacatttctggttggCTTGTAGTTCTTTCTYGGAAAAATATTATTCTAATTTCCTTATAGTATTAGCCATCAAAGTAGGGGAAGCAGATCAAATCTACCATAAGACCAAGTCATAGGAAGAAGAAGATCAAATTAAGATGCTAGGCAAAAGTCTCAGCACATATGGATTATGAGAAGCACATTCACACATCCAAActcaaagaatggactcagcg

For: 5'-3' = cactaccacatttctggttgg

Rev 5'-3' = cgctgagtccattctttgag

M7 = DYS253 (300 bp). C to G at position 236

ActgtgagcgagctgaaaatGCCTGATTTTCTCCCTTGGTTTAATGTAAAGGAAGGGATC CAAAGGCTTAGGGAGATTTGGGATGGTGGATTAGTCACTTTAGACCTACTCAT TCCAATAGGGAGGGTCCAGAAGATGTACCCTTGACCAATGCCTTGCAAAATA GATTCGTGAGGGCAGCACCTGCATCACCAAAGGGCATGTAATCATTCCTCTCT GTATGTCAGATCTAACAASAAGAAGAACAGTAACTCAACTACAAAATTTAAA CACAATGGAAAAtaattggttcacaaggctgc

For: 5'-3' = actgtgagcgagctgaaaat

Rev 5'-3' = gcagccttgtgaaccaatta

M8 = DYS263 (267 bp). G to T at position 137

CccacccacttcagtatgaaTTTTGGGATCTGTTACCTATTTTTTGATATAAAATCAACTG CAAGTTTAGTGCCTCAGTATCACAAACACTGTATTTGCTCATATGTCTGTGAA TCAATAACTTGGACTGGGTTCAKTTGGGCAGTTCTTCTATTGGTCTTGCCTGG GGTCTTTAATGCAGCTTCCATTTTCTGGCAGCTTGATGAGACTGGATGGTCTA AGGTACATTCATGAACACATCTGTTTGgtggacttgtctgtcagcct

For: 5'-3' = cccacccacttcagtatgaa

Rev 5'-3' = aggctgacagacaagtccac

M9 (340 bp) G10.35a C to G substitution at position 68

GcagcatataaaactttcaggACCCTGAAATACAGAACTGCAAAGAAACGGCCTAAGAT GGTTGAATSCTCTTTATTTTTCTTTAATTTAGACATGTTCAAACGTTCAATGTC TTACATACTTAGTTATGTAAGTAAGGTAGCGCTTACTTCATTATGCATTTCAA TACTCAAAAAAAATTCCTTTGTGAAATGTTGAAATATTTTTCTAATCTGTTTC ACGAGCTTCAAAAAATGAGGAAAAAAGATTCAGTTTACATTTCAGCAAAATGC

CTCTTTTTAATCGGATTTATGTTTACTTAACATTTACAGTACATTTACgcttgagcaa agttaggtttt

For: 5'-3' = gcagcatataaaactttcagg

Rev 5'-3' = aaaacctaactttgctcaagc

M10 = G10.10 (343bp). T to C at position 156

For 5'-3 = gcattgctataagttacctgc

Rev 5'-3' = taataaaaattgggtcaccc

M11 = G10.37 (222 p) A to G at position 44.

TctctctgtctgtctctccctccCTCTCTCTTGTATTCTAAC**R**GAAAGGTTTAGAACTTGCA TAATTGGGAAAGAAGCTGTTGCCTGAACTTACTGGGGGATTCAGCATTGTCA TTTTGGACATGTCACTTATCCTCAGTATTTGCTTCCCCCAGGAGAGAGCTGTA ATAAAAAAGCATTGCAATTTAATACATAAgctcagtaagttcttgtttatgctc

For: 5'-3' = tetetetgtetgteteteetee

Rev 5'-3' = gagcataaacaagaacttactgagc

M12=DYS260a (309 bp) G to T at position 286

ATTATTTGTAGAKATGggggtcactatgttgctcag

For: 5'-3' = actanaacaccattagaaacaaagg

Rev 5'-3' = ctgagcaacatagtgacccc

M13 = G10.06 (233 bp) G to C at position 157

TcctaacctggtggtctttcATTGTTTTACAAAGGTGATTTAGTTTTGGGAAGGACTATTC TCCTTTAAACTATAGACTAAATTTTTCTCAAAGTTAGGTTAGTTTATGCCCAG GAATGAACAAGGGCAGTAGGTAGGTTAAGGCCAAGACGGTTASATCTCTCTTTACTGTTATAATTTTCTCATTGTTATATTTTTTGCAAATGTGgttggataaaatca tggctca

For: 5'-3' = tcctaacctggtggtctttc

Rev 5'-3' = tgagccatgattttatccaac

M14 = G10.07 (287 bp) T to C at position 180

For: 5'-3' = agacggttagatcagttctctg Rev 5'-3' = tagataaaagcacattgacacc

M15 = G10.16 (295 bp = ancestral state); derived allele = 9 bp insertion (304 bp) after position 109; Note that there are also two T to G changes immediately before the 9 bp insertion.

AcaaatcctgaacaatcgcCATCACCTATTTGGTGGACGCATAGGCCTGGTCTCTGATCT GGTCGCATGTCCAGAGGGTCTGCTAACCCACTGCACCTAGGGAGACA**TTGTA CAGAGA**CATTGTACCACCTTTTCTCTACTcttcccagactcaacacatttGATTGTATATGC GCATGAGGTAGAAATATAAGATGAAGCAGGGACAGAGTCAACAAGCCAGAA CTAGATGCTTCTACCTGGACAGAAGACCTAGAATTCTTTTTTGGATCCTAAAT TCACCAggaaattttaaccacatgca

For: 5'-3' = acaaatcctgaacaatcgc

Rev 5'-3' = tgcatgtggttaaaatttcc

M15 polymorphic region in more detail

mutant sequence = GACA TT GTACAGAGA CA ancestral sequence = GACA GG ******* CA

M16 = DYS214b (266 bp) C to A

For: 5'-3' = tgttatgtcatttgaacccag

Rev 5'-3' = ccgtgtgttgctgggctgt

M17 = G10.47a (333 bp) -1bp deletion (4G's to 3G's) at position 68
CtggtcataacactggaaatcAGATTCTGTCTACTCACCAGAGTTTGTGGTTGCTGGTTGT
TACGGGGTTTTTTTAAGTGAATTTTGGGGTTTGTTAAGTGGCCAAACTATTTT
TGTGAAGACTGTTGTATGTGGGTTTCAGATGTCTCTACATCAGTTTGTGGTCA
GCTAGTGAGTTAAATTTTATGAAAAAGCCTGGAGAAACAAGAATAGCAGTAAA
AACTTCCAGTCTTTGTAGATTGGGTGTCTTCAGTGCTTAGCTGGGCAATTTAA
AACTTACCTTAAGTAGTACAGTTGGCCCCTTTGTGTCTGTgagtttcacatttgtaggttca

For: 5'-3' = ctggtcataacactggaaatc Rev 5'-3' = tgaacctacaaatgtgaaact

M18 = G10.47b (333 bp = ancestral size) +2 bp (extra AA) insertion after position 62 CtggtcataacactggaaatcAGATTCTGTCTACTCACCAGAGTTTGTGGTTGCTGGTTGT TAAACGGGGTTTTTTTAAGTGAATTTTGGGGTTTGTTAAGTGGCCAAACTATT TTTGTGAAGACTGTTGTATGTGGGTTTCAGATGTCTCTACATCAGTTTGTGGT CAGCTAGTGAGTTAAATTTTATGAAAAGCCTGGAGAAACAAGAATAGCAGTA

$AAAACTTCCAGTCTTTGTAGATTGGGTGTCTTCAGTGCTTAGCTGGGCAATTT\\ AAAACTTACCTTAAGTAGTACAGTTGGCCCTTTGTGTCTGTgagtttcacatttgtaggttc\\$

a

For: 5'-3' = ctggtcataacactggaaatc Rev 5'-3' = tgaacctacaaatgtgaaactc

M19 = G10.47c (333 bp) **T** to **A** at position at 131

ctggtcataacactggaaatcAGATTCTGTCTACTCACCAGAGTTTGTGGTTGCTGGTTGT TACGGGGTTTTTTTAAGTGAATTTTGGGGTTTGTTAAGTGGCCAAACTATTTT TGTGAAGACTGTTGTAWGTGGGTTTCAGATGTCTCTACATCAGTTTGTGGTC AGCTAGTGAGTTAAATTTTATGAAAAGCCTGGAGAAACAAGAATAGCAGTAA AAACTTCCAGTCTTTGTAGATTGGGTGTCTTCAGTGCTTAGCTGGGCAATTTA AAACTTACCTTAAGTAGTACAGTTGGCCCTTTGTGTCTGTgagtttcacatttgtaggttca

For: 5'-3' = etggtcataacactggaaatc Rev 5'-3' = tgaacctacaaatgtgaaactc

M20 = G10.48. (413 bp) A to G at position 118

For: 5'-3' = gattgggtgtcttcagtgct Rev 5'-3' = cacacaacaaggcaccat

M21 = G10.43 (415 bp) A to T at position 357

For: 5'-3' = cttttatttctgactacaggg Rev 5'-3' = aacagcagatttgagcagg

M22 = DYS273 (327 bp) A to G at position 129.

ACATGGGCTGGAGATTCAGTGGGACCCTTTCTTTAGTGTCTGCCTAATGCAAGCTGGCTAACTCCTTTCAAAAGTTTTGTCTTGCTGATgaagcctccaggtagtaggc

For: 5'-3' = agaagggtctgaaagcaggt Rev 5'-3' = gcctactacctggaggctt

M23 = G10.57a (327 bp) A to G at position 159

For: 5'-3' = tetetaacttetgtgagecae

Rev 5'-3' = ggaaaaactaaactctaaatctct

M24 (tetranucleotide TAAA motif) = SRY 8299c. Internal primer regions for SRY4064 which contain M40 and M41.

For: 5'-3' = acagcacattagctggtatgac

Rev 5'-3' = tctctttatggcaagacttacg

M25 = B9.008b. (340 bp) G to C substitution. Position 121

AaagcgagagattcaatccagGATGACAGAATGCGTTCACCTTTAAAGGGATTAAAAGA AGTATAATACAGTCTGTATTATTAGATCACCCAGAGACACACAAAACAAGAA CCGTGAATTSAATTAGTGGTATACTAATAGAGTGGTTTTACCTGAAATATTTA CACATCAATCCTACTGAATTCTTACAACAAATGATTTAGATTAGCTATTGTAT TCACCAGTTGAAAGAACAGAAAATATTGAGGGAGATAACTTGTGTCAGTGCA ACTTAATCAGATTTAGGACACAAAAGCAACTACATAATGAAAAAAGAGAgctggt gacttaacttgctaaaa

For: 5'-3' = aaagcgagagattcaatccag

Rev 5'-3' = ttttagcaagttaagtcaccagc

M26 = B9.005 (321 bp) G to A at position 68

CcagtggtaaagttttattacaatttTTTTAAACCAAGATTCAATTTTTTTCTGAATTAGAATT ATCRCAGAGAACACTGAATGGCCTATGAAATTCAATTTTTGCTGCAGATTTC GTCATGTTTCTTAATGAACATATAACTAACTTCTAATCACAAGATAAATTCTT GCCTATGTGCAAAAACTTAGTGCTGCATCCTTGTGTATGGTTTTAAAAAGTGT

CAAAACTGGCCCCTCATGTCAAATACAGCCCCAATTAGGGGAGGCAACCTAA GAAAGGTGTACAACTGTCCTGACATTggattgcctgcttactgtgaa

For: 5'-3' = ccaglggtaaagttttattacaattt

Rev 5'-3' = ttcacagtaagcaggcaatcc

M27 = G10.65. (526 bp). C to G at position 398.

CggaagtcaaagttatagttactggAAATACAAACTGTGGCAGTAGAAAACCCTAGGCACA
AGGGAAGTAAAATATTAACCACTCCAGGCTGGAGTGCAGTGGCGCAATCTGG
GCTCACAGCAAGCTCTGCCTCCTGGGTTCACACCATTCTCCTGCCTCAGGCTC
CCGAGTAGCAGGGAGTACAGGCACCCGCCACCAGGCCTGGCTAGTTTTTTT
GTATTTTTAGTAGAGATGGGGTTTTACTGTGTTAGCCAGTATGGCCTCGATT
TCCTGACCTCGTGATCCGCCCACGTCAGCCTCCTAAAGTGTGGGGATTACAG
GAGTGAGCCACCATGCCCAGCTGAAACAATAGTTCTTCACAATGGCATCTAC
CACTATGTCCACATTTGCACCTSTGTCCTGAACCTCGATTCCTATAGGTTGAT
GTGTTGAGAACCAGACAATACGAAATAGAAGACAAATCATGAGCTTACAGA
ACCTGAAACTTTTTACACTGGGCAGtgtggtagacagaacagcagtg

For: 5'-3' = cggaagtcaaagttatagttactgg

Rev 5'-3' = cactgetgttctgtctaccaca

M28= G10.33n (332 bp). **T to G** at position 277.

GcttacttgggacacaggctAGTTCTCCTGAAGCTATTGAGCAGTATGTGTTGAGGTG CGCTACGCCAGTTGAGGTGAAGCTGTTACACAGTATGAAAGCCGGGCTTTGT AGCTGCAGCTGCGCATTGCACCCCCAGCTACGCAGTCTCCTTTCCTTCTCAGT CACAGGACCGGATGGCAAGTGGCCGCAGCCAGTCGGTGAGACCGACTGAGC TCTGGGGCTTCAGTTCTTGACGCTACCTACATGGCTACATCTCCAGCCAAGGA TGAGAGGKGATGCCAGAGGACCTCGATCTAAATTGGGCAccattatcgtatgacaacttct ct

For: 5'-3' = gettaettgggacacagget

Rev 5'-3' = agagaagttgtcatacgataatgg

M30 = G10.66 a (486 bp) **G** to A at position 132.

For: 5'-3' = gaaccagacaatacgaaatagaag

Rev 5'-3' = tttageggettateteattace

M31 = G10.66 b (486 bp) G to C at position 71.

GaaccagacaatacgaaatagaagACAAATCATGAGCTTACAGAACCTGAAACTTTTTACACTGGGCAGT\$TGGTAGACAGAACAGCAGTGGCTGCCCAAAGATGATCATGTT

For: 5'-3' = gaaccagacaatacgaaatagaag

Rev 5'-3' = tttageggettateteattace

M32 = G10.68a (370 bp) T to C at position 166.

TtgaaaaaatacagtggaacAAAGATCCTCTGTATCTCTGCTCCTAAGATAGCAGAGACA GCatactggcttctgttcaattttcCTTTGATTACACAACTTCATTGGCTACGGTGTTTAATAT GACCGTCATAGGCTGAGACAAGATCTGTTCAGTTTATCTCAYAAGTTACTAG TTAAATCTCAGACATATTATACTTTTGTAACTGAGTGACTCCCATTGTAAGGA TAACTACTTCAATGTGCGTATAAATGAGTCAGTTGTCTCTCTTGGGGGGCTTCA ACAAATAAGCAAAGATAACCTCATTGTGGAGAGCACTTCACATTTGTTTTTAG GGTTACATAGTCTActctgtatccttaaacacttg

For: 5'-3' = ttgaaaaaatacagtggaac

NewF 5'-3' = atactggcttctgttcaattttc

Rev 5'-3' = caagtgtttaaggatacaga

M33 = G10.68b (370 bp) A to C at position 180.

TtgaaaaatacagtggaacAAAGATCCTCTGTATCTCTGCTCCTAAGATAGCAGAGACA GCatactggcttctgttcaattttcCTTTGATTACACAACTTCATTGGCTACGGTGTTTAATAT GACCGTCATAGGCTGAGACAAGATCTGTTCAGTTTATCTCATAAGTTACTAGT TAMATCTCAGACATATTATACTTTTGTAACTGAGTGACTCCCATTGTAAGGA TAACTACTTCAATGTGCGTATAAATGAGTCAGTTGTCTCTCTTGGGGGGCTTCA ACAAATAAGCAAAGATAACCTCATTGTGGAGAGCACTTCACATTTGTTTTTAG GGTTACATAGTCTActctgtatccttaaacacttg

For: 5'-3' = ttgaaaaatacagtggaac

New F 5'-3' = atactggcttctgttcaattttc

Rev 5'-3' = caagtgtttaaggatacaga

M34 = G10.69 (372 bp) G to T at position 131

CacttcacatttgtttttaggGTTACATAGTCTACTCTGTATCCTTAAACACTTGAAGATCT GTTATAACTACATCTGAGATAGTCACAGTGTTTTCTCATGTTAATGCCTG GCTTCCACCCAGGAGKCACATGTGGTGTGTCTGCAAATAAAGTGTTTATGAT TATTGGGGTCCCCCAAGCTGGACCTGTATCCATGTTCAAGTGGCCACAGGGTT ACTTGCTTTAGCATGGCTCCTTGGCTGGCTGTTAAGTGAATAATTAAACTGAG TCTTTTTTGCAGGAGCTAACTGAGACCAATCAATCAGTCAATTTTCCCTTTCT GTGTGTAACACAAGCTGGATGTCCctggaatgactaaataatgact

For 3'-5' = cacttcacatttgtttttagg

Rev 5'-3' = agtcattatttagtcattccag

M35 = G10.72a (514 bp) G to C at position 168

For: 5'-3' = taagcctaaagagcagtcagag

Rev 5'-3' = cagaggagcaatgaggaca

M36 = G10.82a (436 bp) T to G at position 74

AgatcatcccaaaacaatcataaCTTGTTTAAATTGTTCATAGCAAAAAGTTACATATTATA AAGAGTTATGAG**K**GTCTTAGGCAGTGAATAGTAACTGAATATCCTTTTATAG TTGTCCTTCACTAGCAGGAAGCCTTATTCCCTGCCCTTTTACATATCTTAACTT AGAATGTTACTGTCTAAATAGTGGTTAGGCAAGAGTAGTTCTTAAACGTGCA GTAATTATCTTGCACTACATTTAAGGGCTAAATAGCTAGTAGTGGTGCTTGAT AATTGAAGAAATTTGTACAGCTGGAGGAAGTACCTGCTAAATTTCAAAAGT TACCTGAATTTAATAGGTAAATCTGTTTTTAATTAGAGCTATATCATTTACTC TGAATGTCTTAACATAGAAGTTTACATAAAATTTACAGattggattgatttcagcctt

For: 5'-3' = agatcatcccaaaacaatcataa

Rev 5'-3' = aaggetgaaatcaatccaatctg

M37 = G10.STS 84 (422 bp) C to T at position 203. This STS also contains M61 at position 101 which is defined in G10.83.

For: 5'-3' = cagattggattgatttcagcctt

Rev 5'-3' = agcatacaaaaaaaaaaaaactgc

M38 = G10.73a (337 bp) T to G at position 146

Rev 5'-3' = ttaaagaaaagaaaagcagatg

M39 = G10.73a (337 bp) - 1 bp (-C) deletion at position 236

For: 5'-3' = cagtttttagagaataatgtcct

Rev 5'-3' = ttaaagaaaagaaaagcagatg

M41 = SRY 4064b (218 bp) G to T at position 117. Site is located within SRY 8299 509 bp STS.

GtataataggctgggtgctgTGGGTCACACCTGTAATCCCAGCCCTTCGAGAGGTCAAGG CAAGCGGATCACAGGGTGGAAGAGATTGAGACCATCCTGGCCAACATGGTG AAACT**K**GGTCTCTACTAAAAATACAAAAAATTAGCTGGGCGTGGTGACATGT GCCTGTAATCCCAGTTACTCGGGAGGCTGAGGCAGaagaatcatttgaactcatg

For: 5'-3' = gtataataggctgggtgctg

Rev 5'-3' = catgagttcaaatgattctt

M42 = B9.008a (340 bp) A to T substitution at position 297

For: 5'-3' = aaagcgagagattcaatccag Rev 5'-3' = ttttagcaagttaagtcaccagc

M43 = DYS260b (309 bp) A to G at position 77

For: 5'-3' = actaaaacaccattagaaacaaagg

Rev 5'-3' = ctgagcaacatagtgacccc

M44 = G10.87 (389 bp) G to C at position 263

CCTGAGGAAACTCATGCGAGAAATGCCAGAAAAAGAAGACAGCAACAAAGA AGATAAAAGAAAGACTGACAAAAGCATTGAATTTCTGGTAGAAAAASCAGT GTACTAGAAGGTTAGGAGATTTCCTAGCTGTCAGCCATGAAAGGGTTGGGGA AGAAAGAGCAATTTGGTTGCATACTGTAGCATGGTCATCTAGGGTGgtcctcaaac acatagaaatcaca

For: 5'-3' = ctggcaccttctgatattttgag Rev 5'-3' = tgtgatttctatgtgtttgaggac

M45= B9. 12(352 bp) G to A substitution at position 109

For: 5'-3' = gctggcaagacacttctgag Rev 5'-3' = aatatgttcctgacaccttcc

M47 = G10.82b (436 bp) G to A at position 395

AgateateceaaaacaateataaCTTGTTTAAATTGTTCATAGCAAAAGTTACATATTATA AAGAGTTATGAGTGTCTTAGGCAGTGAATAGCAAAAGTTACCTTTTATAGT TGTCCTTCACTAGCAGGAAGCCTTATTCCCTGCCCTTTTACATATCTTAACTTA GAATGTTACTGTCTAAATAGTGGTTAGGCAAGAGTAGTTCTTAAACGTGCAG TAATTATCTTGCACTACATTTAAGGGCTAAATAGCTAGTAGTGGTGCTTGATA ATTGAAGAAATTTGTACAGCTGGAGGAAGTACCTGCTAAATTTTCAAAAAGTT ACCTGAATTTAATAGGTAAATCTGTTTTTAATTAGAGCTATATCATTTTACTCT GAATGTCTTAACATARAAGTTTACATAAAATTTACAGAGTAGTGGTGCTt

For 5'-3'= agatcateceaaaacaatcataa

Rev 5'-3' = aaggctgaaatcaatccaatctg

M48 = G10.79n (240 bp). A to G at position 160

For: 5'-3' = aaacaatatgtatgctaattttgct Rev 5'-3' = tcaatgtaaatgttagtataaggatg

M49 = B9.15new a (354 bp) **T to C** at position 229

CggcaacagtgaggacagtAGCTCCAGGTCTGGGCGGAAGGTGGTGCGGTGAAAGGTG CAGGGACAGACTGGGTTAGAGGCCACTCTTGGTCTTATCCTCCATGGCCACA ACAGAGGTGACAAATACATGGGTCACTCAGTTATGTTTAGCCAACAGCCTAC CCAAACCACACCTGTCTTACCAGAGCCCTTTCCTGGAGCCATGTTCTCAGGAC TGGTCACACTGTCYCCATTCTCCAGCAGCCCTTTGGACCTATCGGAAAAAAAAG

AATGGGTAACAATAATTGAGCTGATGAACCAGGTCCTATCTTTCCTCCACAA CTCCAAAACTTGGgagcctctatctcctgaagca

For 5'-3'= cggcaacagtgaggacagt

Rev 5'-3' = tgcttcaggagatagaggctc

M50 = B9.15new b (354 bp) **T** to **C** at position 175

For: 5'-3' = cggcaacagtgaggacagt

Rev 5'-3' = tgcttcaggagatagaggctc

M51 = B9.16 (339 bp) G to A at position 33

For: 5'-3' = gagcetetateteetgaage

Rev 5'-3' = tgactgctctgttgcgaca

M52 = G10.88 (534 bp) A to C at position 477

For: 5'-3' = actgtagcatggtcatctagggtg

Rev 5'-3' = gacgaagcaaacatttcaagagag

M53 in tree (tetranucleotide TAAA motif) = SRY 8299d. Internal primer regions for SRY4064 which contain M40 and M41.

AcagcacattagctggtatgacAGGGGAGATGTGATTAATTGACCTACTGATAAGACTCA TTTCAGTAAATGCCACACAAGAATgtataataggctgggtgctgTGGGTCACACCTGTAA TCCCAGCCCTTCGAGAGGTCAAGGCGAGCGGATCACAGGGTGGAAGAGATT

For: 5'-3' = acagcacattagctggtatgac Rev 5'-3' = tctctttatggcaagacttacg

M54 = B9.17 (360 bp) G to A at position 164

For: 5'-3' = cctcctctggtctgggttt Rev 5'-3' = tgtgtcaggactggttccat

M55 = B9.28 (382 bp) T to C at position 228

For: 5'-3' = cgtaggcgtttgacagcag Rev 5'-3' = cctttcttcgtaatcctccc

M56 = B9.29 (399 bp) A to T at position 39

CcagaaactgaagtacaaatgcAATGGGAGGATTACGAWGAAAGGAGGCTAAGTGAT GATAAGTATGGTCAGAATAAATAAATTTATTCTAGACAAGAAATGAGAGTTCA TTATGTCAGAAGCAAAATAGTACTACAGGATGACAACTTCTGAGATTTACTCT TTGGTTCCAACTGCCTACAAGACAAAGAAAACTGAAGAGGCCAGGAAGTTAA ATGCATGAGGAAAACTTGAGGCAGATTAAAATGGAAATGCAGGGCATGTTAT TTGGGTATCATGGGTTCAATCTGGAAAAGCCTTATTTCTCCTGAACCACAGTA GGGAAAGGAGTTATCCAGAAAAGTGAAATTTATCTAAAATTTTAAGTTTCC ATGTTTTaaagagaggcagcaatgaga

For: 5'-3' = ccagaaactgaagtacaaatgc

Rev 5'-3' = tctcattgctgcctctcttt

M57 = G10.85n (326 bp ancestral); +1 bp insertion (327 bp = Derived). Extra A inserted at position 133

For: 5'-3' = attgggaggaagtggtttctg Rev 5'-3' = gttctgtatcttttccattatttgc

M58 = G10.57b (327 bp) G to A at position 224

For: 5'-3' = tctctaacttctgtgagccac Rev 5'-3' = ggaaaaactaaactctaaatctct

M59 = B9.15new c (354 bp) A to C at position 279

For: 5'-3' = cggcaacagtgaggacagt Rev 5'-3' = tgcttcaggagatagaggctc

M60 = B9.34 (388 bp ancestral); **+1 bp insertion** (389 bp = DERIVED). Extra T inserted after position 242

GeactggegtteateatetGGGAGCAGCTCAAAAGCCTCTCGCTCAGCCTCCGTGACGCC CTGGGGGTGTTCAACCCACATATACTGTAAAGACTAGGAGTAGGGTTGTGGA CACCCACCTCAGCCAACACTGAGCCCTGATGTGGACTCAACCTTGTAAGGA AAGCTGTAGAGAAATTGGAAGAAAAAATATAAACACATACAGACTCTGTCTT TACATTTCAAAAATGCATGACTTAAAGTATCAGGCACACAGTGGTTACTCAAT GTTGGTCTGTGTCTCTGTAACGTAATATATGTGACTAAATCCCTAAGCTCTGC TCTTGACCACCCACCTTCTCCAAAAGGGCCTTTCGTAGACGTCGCTcctcctgaacca taatgaacat

For: 5'-3' = gcactggcgttcatcatct Rev 5'-3' = atgttcattatggttcaggagg

M61 = G10.83 new a (190 bp) C to T at position 98.

For: 5'-3' = attggattgatttcagccttc Rev 5'-3' = attttattttctgtgttccttgc

M62=DYS260c (309 bp) T to C at position 60

For: 5'-3' = actanaacaccattagaaacaaagg

Rev 5'-3' = ctgagcaacatagtgacccc

M63 = B9. 22 (308 bp) G to A at position 43

For: 5'-3' = ctcttcccttggttcctattc Rev 5'-3' = ttaggcagaagcatccacc

M64 = B9.t23 (325 bp) A to G at position 279 RECURRENT

TatagaccetgactactcaagagaaAAGTCCAATCCAAAGAAAAAATACAAAAGAAAACA
AAATCACATCAGGCCACAAACCAGTTTAAGGGCCCTCACCACATGGTTGGCT
CCAGACTGAAACATTTCATAGGGGTAAATAATGCGTTCGTAATGTGATCGTA
GCAGGGAGCCAATGTTTTTTGCCTGGTGGGTAGTGGAGACGCTGGGCAACTCG
AGCCCACCGACGATCCTTGCAGATGGCTTCATAGCCACCTTCCTCAATCACAA
TCTGAAAGTRTAAGAAACAATATGGATGAACTGTGAacagactggaaagggctacc

For: 5'-3' = tatagaccctgactactcaagagaa

Rev 5'-3' = ggtagccctttccagtctgt

M65 = B9.t26 (436 bp) A to T at position 152

TtctgatgccagcttgttcgGGTCAGAAAAGTTAAATGAGAAAATTTGGTGCTAAGGGTTTCTGGTCATGAGTGTAAATAACGCCTCGCCAAGTGGTAAACTGCCCCAACGTTCAAACCAAAGGCTACCCATTCCCAAATTTTGTTTCAAAGWCTTACCGCGGGTGGGCGGATTTTGCAGATGCCAGACTTCTCTGCTATGGGCCTTATTTTCGCAATGTAGCCAAGCGGGTCTTGGAATTCAGCCCAGCTAGGCTCAAAAACCGGGCACTCCGGTGGCGCGCAGGAACTCGTCACACCCCGGTTCCATGTCGGGCCTTAATGCTAAAGCTGTAAAAATAAGAATCACATTGTCTTTAATGACGCGCTGGTTCCTCCTACTAAAAAGGCCTATGAAAAATTTCATTTTCTTGAGAATTTCaaggttactttaatcccgtagc

For: 5'-3' = ttctgatgccagcttgttcg Rev 5'-3' = gctacgggattaaagtaaccttg

M66 = B9.41 (415 bp) A to C at position 135

For: 5'-3' = ctgtgtaacaccatcaagtgc Rev 5'-3' = acatcttctggagacatacttcc

M67 old = B9.36new a (409 bp) **A to T** at position 377

CcatattetttatactttctacctgcAGGCCCACTGCATGCTCACTCACCCAGTCAGCAGTACA AAAGTTGACAGCTTCAGCAAAATTGTAGCCTTGGTTAAAACCACTGTGGTAA GCACGAGGAAAAAGTGATGACAAACTCCCCTGCACACTGGTTTGTGCGGACAA CCTAAAAAGGAGAAAAAAGCAGAAAAGAGGTGTGGGTCAGAACTAATGGGCC AGATGTGAACTCAAAGATGTCTCTAGATGCTGTAACAGATGTAGGAAGAGTG GAAAGGCTCTATCTTCAAGTACGTGTCCTAAAAGAAAAATGAGATTGTGAAT TTAAAAGTGGTATTCATAGAAAAAGTACTCAAAAATATGTGTAATTCAAAAAAAC

 $\mathbf{A}\mathbf{W}\mathbf{A}\mathbf{T}\mathbf{A}\mathbf{T}\mathbf{A}\mathbf{G}\mathbf{A}\mathbf{G}\mathbf{G}\mathbf{G}\mathbf{g}\mathbf{t}\mathbf{c}\mathbf{c}\mathbf{a}\mathbf{c}\mathbf{g}\mathbf{a}\mathbf{a}\mathbf{c}\mathbf{a}\mathbf{g}\mathbf{t}\mathbf{g}\mathbf{a}\mathbf{a}\mathbf{a}\mathbf{g}\mathbf{a}\mathbf{c}$

For: 5'-3' = ccatattetttataetttetaeetge Rev 5'-3' = gtetttteaettgttegtggae

M67 revised B9.36new a (386 bp) STS A to T at position 327

CCAgtcagcagtacaaaagttgACAGCTTCAGCAAAATTGTAGCCTTGGTTAAAAACCACTG TGGTAAGCACGAGGAAAAGTGATGACAAACTCCCCTGCACACTGGTTTGTGC GGACAACCTAAAAAGGAGAAAAAAGCAGAAAGAGGTGTGGGTCAGAACTAA TGGGCCAGATGTGAACTCAAAGATGTCTCTAGATGCTGTAACAGATGTAGGA AGAGTGGAAAGGCTCTATCTTCAAGTACGTGTCCTAAAAGAAAATGAGATTG TGAATTTAAAAGTGGTATTCATAGAAAAGTACTCAAAATATGTGTAATTCAA

newFor 5'-3' = c:cagtcagcagtacaaaagttg newRev 5'-3' = g:cattctttgattatagaagcaa

M68 old = B9.36new b (409 bp) A to G at position 268

CcatattctttatactttctacctgcAGGCCCACTGCATGCTCACTCACCCAGTCAGCAGTACA AAAGTTGACAGCTTCAGCAAAATTGTAGCCTTGGTTAAAACCACTGTGGTAA GCACGAGGAAAAGTGATGACAAAACTCCCCTGCACACTGGTTTGTGCGGACAA CCTAAAAAGGAGAAAAAAAGCAGAAAAGGGTGTGGGTCAGAACTAATGGGCC AGATGTGAACTCAAAAGATGTCTCTAGATGCTGTAACAGATGTAGGAAGRGTGTGAATGTGAAT

TTAAAAGTGGTATTCATAGAAAAGTACTCAAAATATGTGTAATTCAAAAAAC AAATATAGAGGGgtccacgaacaagtgaaaagac

For: 5'-3' = ccatattetttataetttetaeetge

Rev 5'-3' = gtcttttcacttgttcgtggac

M68 revised B9.36new b (386 bp) STS A to G at position 219

ccagtcagcagtacaaaagttgACAGCTTCAGCAAAATTGTAGCCTTGGTTAAAAACCACTG TGGTAAGCACGAGGAAAAGTGATGACAAACTCCCCTGCACACTGGTTTGTGC GGACAACCTAAAAAGGAGAAAAAAGCAGAAAGAGGTGTGGGTCAGAACTAA TGGGCCAGATGTGAACTCAAAGATGTCTCTAGATGCTGTAACAGATGTAGGA AGRGTGGAAAGGCTCTATCTTCAAGTACGTGTCCTAAAAGAAAATGAGATTG TGAATTTAAAAGTGGTATTCATAGAAAAGTACTCAAAATATGTGTAATTCAA AAAACAAATATAGAGGGGTCCACGAACAAGTGAAAAGACTCTttgcttctataatcaaa gaaatgc

newFor 5'-3' = ccagtcagcagtacaaaagttg

newRev 5'-3' = gcatttctttgattatagaagcaa

M69 = B9.62a (2.57 bp) **T to C** at position 222

GgttatcatagcccactatactttgGACTCATGTCTCCATGAGAACTAAGACTACCACAACA GAATCCCTATAGTCCAGCCCTCAGATCACATACATGTACAGGCATGTTGAAG TAGTCGGACTTGAAGGAATCAGCCATTTCACCAAAACTCTGCAAACTGTACT CCTGGGTAGCCTGTTCAAATCCAAAAGCTTCAGGAGGCTGTTTACACTCCTGA

AAYAAATATATTTCagcaagacaaagggaataaagat

For: 5'-3' = ggttatcatagcccactatactttg

Rev 5'-3' = atetttatteeetttgtettget

M70 = B9.62b (257 bp) A to C at position 45

GgttatcatagcccactatactttgGACTCATGTCTCCATGAGA**M**CTAAGACTACCACAACA GAATCCCTATAGTCCAGCCCTCAGATCACATACATGTACAGGCATGTTGAAG TAGTCGGACTTGAAGGAATCAGCCATTTCACCAAAACTCTGCAAACTGTACT CCTGGGTAGCCTGTTCAAATCCAAAAGCTTCAGGAGGCTGTTTACACTCCTGA AATAAAATATATTTCagcaagacaaagggaataaagat

For: 5'-3' = ggttatcatagcccactatactttg

Rev 5'-3' = atctttattccctttgtcttgct

M71 = B9.63b (328 bp) C to T at position 197

TtgaattatagtcccttgcctcTGGTTCAGTCAAGTCTCTATCATTCTAGAGTTAGTGTGTT CAATCGTTCTTGTATAGTAGCTCACTGATAGCTTAATCAAAACCTAACACAAA TATTAACTTATAAAAAGGGCAGAAACTACCTTCCCAAAACCCAGAAGGGGAGA TTACAGAAAATCACCAACCAAAAATAAAGYATCTGTGACAGACAGATCTTAC CGCCAAGATACATTTTGGGCACCTCCAGATGCCTCTGGGGATTTCAGGAAGG GGTGGTAACAAGCAGAAGATGTGGTAATTGTCATCAcagccatcacagaaaagaagc

For: 5'-3' = ttgaattatagtcccttgcctc

Rev 5'-3' = gcttcttttctgtgatggctg

M72 = B9.63a (328) A to G at position 157

For: 5'-3' = ttgaattatagtcccttgcctc

Rev 5'-3' = gcttcttttctgtgatggctg

M73 = B9.47a (361 bp ancestral & 359 bp derived) -2bp deletion,

(-GT) at position 260

cagaataataggagaatttttggtCAAATAAAAGGCCATATTATATTTCTTTTGATAAAAGT ATCATGTGTTCAGTATGTTTTATTATTTGAAATAATAACATGACAGGAATAT ATTTGAAAAAAATTCCAAAAAAAGCTAAATATACAAACTAAGAAAAATTATAT GATTATACTTATCTGCAGTATTGTAAAAACAATAGTTCCAAAAAACTTCTGAATT ACAAGTTTAATACATACAACTTCAACTTCAACTACATT**GT**GGTTAGACGTTC AGAGGAATCACAAAGGACCTCAACATGCTAGATAAGAAAAATGTATTTTTAA ATGTTTTGGCTCAgctgcttagaaaataaggaaaat

For: 5'-3' = cagaataataggagaatttttggt

Rev 5'-3' = attttccttattttctaagcagc

M74 = B9.50a (385 bp) G to A at position 195.

For: 5'-3' = atgctataataactaggtgttgaag

Rev 5'-3' = aattcagcttttaccacttctgaa

M75 = B9.51 (355 bp) G to A at position 296

GctaacaggagaaataaattacagacTGTAAAAGTTGATGACCAAGAATTTTTCAGAAGTGG TAAAAGCTGAATTCTCAAGTTTGAGAATTCCTATCTATTCCCAGAAATATTAA GTAAAAAGTCACATTCCACACATCAAGAAAACTTGCAAGACACTAAAAGAG ATATTATAGCAGTCAAATAGAAAAAGCAAAATAGACTACTACAAATTAATGT AAGATTCAGAATTGACTTGTCAAAAGCCAAAACAGATTTCTAATGTACTGTG AAAAGACAATTATCAAACCACATCC**R**TATATATACAGAGAAATACCTTTATA AGAATAAAAATtcacaaatgcctctgttcaata

For: 5'-3' = gctaacaggagaaataaattacagac

Rev 5'-3' = tattgaacagaggcatttgtga

M76 = G10.100a (493 bp) T to G at position 339

TagaagtagcagattgggagaggACATGTGTTCAAGTTGTACTACTTGTATGTCTTGTTTA
GATATTACAGTCTTTTTCTTTTATCAGAAAAATAATTGAATAATGATAAAATCA
GTTGCAGATTAAGACAGATTATCTGTTGCAGTCTTCTCAAAAACTTAATTTAAG
TACATTATTTTCAGCTAGCATTTCTTCCTTCACATAGAACCTCCATGTGTGGA
GGGATTTCCTAATGAGTCTATTGTATGTACAATAGCACTTAATGACATAGCTT
TTAAATAATAACAGGATTTTACCAAATGTTTAATATGTGCCAGGCATCAAGC
ACCTTACACAGTTKAATTATTGCATAGATTTGGACAGCAACTCTGCAAGTTA
GGTATGGTCATGAACCTTTGCAGATAAGGAAACTGTGTTTCACAAGGAGAAG
AAATTGTCCTGGATCATACAATAAGCTAGGATTTGCTCCAgaccatttttttcattttatcagg

For: 5'-3' = tagaagtagcagattgggagagg

Rev 5'-3' = cctgataaaatgaaaaaatggtc

M77 = G10.105 (371 bp) C to T at position 129

CttttettecettagetgitceTTTCCTGTGGTTTTAAAAAAAGTGACCAGAAACTAGGTCTCTATTTTCATTGCTTTTGCTGCATATTCTTTTAACCTGCTTTTATCTTTTACAGAGTTGAGGGGGCTTTYTAAAATAACCTAGACAATGTCAAGATTCTTAGCTGCGTTTTCTGTCTAAAAGTGTAGATGTCTAGTTATTCCTCATGTAAAACACAACATTTCAACCCTGAGTACTATAAACTTTATTATGCTTCTAGGTTACTTTTTCTCTTTAAGCAATTTATTCCTACATTCCTACATTCCTAAGTGTTCACCAGTGGAACAGATAAGAGATAGAAGTAGTTAGAAATTGAGATAATTGGgttgacctgtcattgttgc

For: 5'-3' = cttttcttcccttagctgttcc Rev 5'-3' = gcaacaatgacaggtcaacc

M78 = B9.60a (301 bp) C to T at position 197

CttcaggcattattttttttggtTCTCCACTACAGGAGAAATGTAAATGTGATGAGTCAGAAT TTAGGATGGCTGTATGGGTTTCTTTGACTAATACAAGAAATCACTTTGTAATG AATGAAATCAGTGGTTTCTGCATTACTCCGTATGTTCGACATGAACACAAATT GATACACTTAACAAAGATACTTCTTTCYGCCCTTCCAAATATTTCAAAATAAG CTGGTCATAGTACTTGCTTTTCATAAAAAAGATGGTAAGCTTCCAATATTTAGA TTTaaggaaaggtgaaggaacactat

For: 5'-3' = cttcaggcattatttttttggt

Rev 5'-3' = atagtgttccttcacctttcctt

M79 = B9.42 Homopolymer in tree (425 bp = majority men). A's. 8 A's to 9 A's (426 bp derived). Extra "A" inserted after position 212.

For: 5'-3' = agccagttggatgacacgtt

Rev 5'-3' = cctcttcgtttcctgacattt

M80 = G10.107. **Homopolymer in tree** (290 bp = most men). 9 T's to 10 T's (291 bp derived). Extra "T" inserted after position 55.

For: 5'-3' = acttletettettttagggtgace Rev 5'-3' = acaeacacacacaaatettatgg

M81 = B9.58a (422bp) C to T at position 147.

ActtaatttatagtttcaatccctcaGTAATTTTAACTTACTTCTATTTTAAGAACTATAACCA AACTATCTGTAAGACTTTTAAGCACTATCATACTCAGCTACACACTCTCTTAAC AAAAGAGGTAAATTTTGTCCTTTTTTGAAYGTCATAGAGTATACTCACACAA ACCAAGAAGAAACAATCTACTACATACCTACGCTATATGGTATATAACTATT GCTCCTAGGCTACAAATTAGTGCGACACTATTGTACTGAATATTATAGGCCAT GTAACACAATGGTTTAAGTATCTGTGCCTCTAAACACAGAAAAGATATAGTG AAAGTACAGTATTGCTCCTTTATTAAACTCAAAATGTTATGCAGCATATGACC GACTATAAAATAGCGCTTATCcagatacagacatctccatgaa

For: 5'-3' = acttaatttatagtttcaatccctca Rev 5'-3' = ttcatggagatgtctgtatctgg

For: 5'-3' = ctgtactcctgggtagcctgt Rev 5'-3' = aagaacgattgaacacataactc

M83 = B9. Alu01 (503 bp) C to T at position 120

For: 5'-3' = gggaaaggagttatccagaaa Rev 5'-3' = aatgacccatgcttacttagc

M84 = B9.72 Homopolymer in tree(439 bp = most men). 9 T's to 8 T's (438 bp derived). One deleted "T" at position 400.

For: 5'-3' = ccctctccaactgagttcaag Rev 5'-3' = gcaatatacgtttctgcttcca

M85 = B9.67a (568 bp) C to A at position 437

For: 5'-3' = aacagaattatcaggaaaaggttt Rev 5'-3' = gcaatatacgtttctgcttcca

M86 = B9.t25a (324 bp) T to G at position 85

TCCAAGTGGTTAACACACAAGCKTATATAACTTGCTTCTGTCATAGATCAAG
TCCAAGTGGTTAACACACAAGCKTATATAACTTGCTTCTGTCATAGATCAAG
TACTTCTGAGTAAGCTATTTTTTTTGCGGTTAAATGTAATAAAAGCTTGTGTAT
GCCTAAACTATATTTAATAACAGCAGAACGTAGAAATATTTGAATCTTATATT
TTTGTCCCTACAGCAGTCAGATGTTTAGAACCCCGTGGAATGTGGCGATCTGA
TACTAATATTCTGATGCCAGCTTGTTCgggtcagaaaagttaaatgagaaa

For: 5'-3' = teccattatttgetatatttget Rev 5'-3' = ttteteatttaaettttetgaeee

M87 = B9.t25b (324 bp) T to C at position 277

TcccattatttgctatatttgctACATACATCTAAGGTCATATCAAAGAAAGAAAACACCAG TCCAAGTGGTTAACACACACAGCTTATATAACTTGCTTCTGTCATAGATCAAGT ACTTCTGAGTAAGCTATTTTTTTGCGGTTAAATGTAATAAAAGCTTGTGTATG CCTAAACTATATTTAATAACAGCAGAACGTAGAAATATTTGAATCTTATATTT

TTGTCCCTACAGCAGTCAGATGTTTAGAACCCCGTGGAATGTGGCGATCTGAT AC**Y**AATATTCTGATGCCAGCTTGTTCgggtcagaaaagttaaatgagaaa

For: 5'-3' = teccattatttgetatatttget Rev 5'-3' = ttteteatttaaettttetgaeee

M88 = B9.80 (314 bp) A to G at position 166

AttetagggtcaggcaactaggGAATACTGCTGTAGCCTAGAGCCTGCCAAAATTATTCA AACTAGCCAATCCCATACTTCTTATCCTGCTCTTGTCTTGCCTTTCCCTTGGTAA ACCCAATATAGGCTATGGCCTAGGTGCTTTTCTTATTCCTGCTTCTTCTGCRT ATCCAAGATAGGTTTTCCTCTCTAGCACTGTGTAGCATATAGTGACTACCTCT CTAAGGCCTGTGATAATAATAAAACTTTGCTTTCCTGAGTCTCTGTGGTCACAC CTACTGACCATCACATGgaagaccatagaatagaacaaaca

For: 5'-3' = attetagggtcaggcaactagg Rev 5'-3' = tgtttgttctattctatggtcttcc

M89= B9.94 (527 bp) **C** to **T** at position 347

For: 5'-3' = agaa.gcagattgatgtcccact Rev 5'-3' = tccagttaggagatcccctca

M90 = B9.96 (331 bp) C to G at position 170

TgatgtttcttcagtctttgaggTTGCTGTCTTTTGGATTTTTGAAAAAATCCTATTTAATAA CTTAGTGGGTTGGTTTGTAGCAACAGTGAATTCAATCAACTGGCTTTATTTCT AGAATATTTTAAAGATATTTTATCTCAGGATTTCTGGATGGTGTTCTGTAACT STAGGGACTGGGAATGAGCTTTGGCTTTGTTCCTTTACACCCTGAGGTTAGAA ATCTGCTGCACTGGAGGGACCAAGATGCTCTCAGAGAAAATGGTCACAACACT CTAATGATTGGTAGTAGCCAATGTGCTTCATATGCGggtggtagcaggattcatctt

For: 5'-3' = tgatgtttettcagtetttgagg Rev 5'-3' = aagatgaatcetgetaccacc

M91 = B9.87a Homopolymer.(495 bp, most men = 9 T's). Either one T deleted or inserted at position 368 (i.e. 8 T's or 10 T's)

GagettggactttaggacggGGAAAAGAAGTGCTAAATGTTTTTGAATAAAACCTTTACT GCACATGATAAACATCCCTTAAAAATTACCTAGGAGCACCCTAAATTTAAA ATGATCACAAAGACCTGGACAGATTACAGTAAACCTTCAACATCGCTAAACA CACGTACCATAAATCAAAAGAAACACACTGCTAATGATCCGTTTTTTGATGT GGAAATATCATGCTGTTTTTAAGGGAAATTATACTTTATTGCGATGTTTTATT TCAAAACAAGATGTTACACTTTATTTCCTATAATTTTATTACAATATTTTACA CCCGTTAAGCAAAAATCCCCCTACATTGCTATTCTG**TTTTTTTT**AATCAG TTCACTACTGTAGTATCTTTTTGTTCTCCATATATTTTTGAAAAAATACGCAAAA GGTAAGTTTTAAAAATCAAATGGTAGATTTTATTTGGAAGGGCACTgccagaagtg ccttaaagttt

For: 5'-3' = gagcttggactttaggacgg Rev 5'-3' = aaactttaaggcacttctggc

M92 = B9.G2 (470 bp) T to C at position 340

TtgaatttcccagaattttgcAATCTGATCCAAATAGTTCAATTTCACTCTAGTTTGGGCCT GGGAAAGAGGGGCCTTATAAGATTGGCATACTCCTTAACCTGACTTCATCG AGTATGCAGTAAATGAACAAGTATTATTCTATGCTATCTACACTTCTCCACCA ACGTGCCGGAGCCCCAGCTTCACTGTCTTATCTCACCAGCGGGGTCCACAAA AAGCTCAAATAAGCTGAGTCTTTAATCTATAAAGAGCTAAGAATGTGCCGTC TTAGGATCAACATCATGTCTAAATTTAAGGAATTATTCTTGGACTTAAAGGTG GCTTGACCAAAAAATAYGTAGGCTCCAACAGTATTTAGACTCAATATCATCAA GACACTCATTTAGAATGTACTGATATATAATTCAAAGAATTAAAATATTTTTC TAGTTCATGTAAAAGAGCTggacacaaaaccagtttctgaa

For: 5'-3' = ttgaatttcccagaattttgc Rev 5'-3' = ttcagaaactggttttgtgtcc

M93 = B9.93 (504 bp) C to T at position 459

For: 5'-3' = aacaaaacaaaacaaaatactgaa Rev 5'-3' = ggttcacttgaagatagtttaggtta

M94 = B9.122 (405 bp) C to A at position 227

For: 5'-3' = cacatggagaacagagaaatgc Rev 5'-3' = cttgtgaaatgttgtgaaagtgg M95 = B9.123 (480 bp) C to T at position 172

For: 5'-3' = gagtggaaatcaagatgccaag Rev 5'-3' = gggctttctgtaacctggaga

M96 = G3.05a (440 bp) G to C at position 70. Internal lower case denotes location of alternative reverse primer region to amplify site a only, as 212 bp STS.

GttgccctctcacagagcacTTTAAAGTGAGCTGTGATGTGTAACTTGGAAAACAGGTCT

For: 5'-3' = gttgccctctcacagagcac Rev 5'-3' = aagglcactggaaggattgc

M97 = G3.05b (440 bp) T to G at position 355

gttgccctctcacagagcacTTTAAAGTGAGCTGTGATGTGTAACTTGGAAAACAGGTCT CTCATAATAGGATAAAACACTCAGGTATAATATTAAAAACCTATGGCAAAAT ATATGGTCCTTTACAAAGCAACAAAGTGGGTGGGTGAATCTCTTCATTCTTGG CTGGCCATCAGTTCCTGTTACTGTACAGGAGTGGGAAAACAGTAGCCCTGGG AAATGGGTTAAAACTGAGTAGGCATCTCCTGTGTCCAATAAGAACTCAATAT TTTTGTCTGCTATATCAAGGGTTACTTGAGGCTCCTCTGTGGAGATGGTAAGT TGTCCAGTGGGAGATATAGAGAATGTTAGGCCKTATAGGTTCTCTACTTTTTT GGCCATTATGAGTCTGAATGTCTCAAACTCCCTTTTTATCCTGGTgcaatccttccagt gacctt

For: 5'-3' = gttgccctctcacagagcac Rev 5'-3' = aaggtcactggaaggattgc

ATATCTGTGTAAAGGCAAGTATTTGAAGCTAGGAGAACTGTTCCTTCTGGCCT GTTGCCCTCTCACAGAGCACTTTAAAGTGAGCTGTGATGTGTAACTTggaaaacag gtctctcataatagg

For: 5'-3' = gaatggggtgttacatggaga Rev 5'-3' = cctattatgagagacctgttttcc

M99 = G3.04b (395 bp nominal) 1 bp deletion (3A's to 2A's) at position interval 96-98, STS alos has polymorphic (GTTTT) motif

For: 5'-3' = gaatggggtgttacatggaga Rev 5'-3' = cctattatgagagacctgttttcc

M100 = G3.04c (395 bp nominal) in tree (penta microsatellite) (GTTTT)5; (GTTTT)6 = most men); (GTTTT)7; (GTTTT)8 alleles detected

For: 5'-3' = gaatggggtgttacatggaga Rev 5'-3' = cctattatgagagacctgttttcc

M101 = A8.05a original (460 bp) C to T at position 154

For: 5'-3' = tcacagcagcttcagcaaa

new R 5'-3' = ataaaaattagactctgtgttactagc3' (used with F primer, just amplifies (369 bp) the first 2 sites including homopolymer T region

Rev 5'-3' = cctttttggatcatggttctt

M102= B9.101 (480 bp) G to C at position 301

AaactgggacacttgtaatgaatAATTACTTTGTTTGTAAATCACAATAGAGATTCTCCATA
TCAAAGCTGTGAACTGTATTCTATAGTATTTAGGCAAATAAGATAGCTACAA
ATTTAAGTACTGTAATAATAGATGCCTGACAATATGTGCTATAGGTAAATCTT
TGAAATTTAATTAAATGAAGTATAGATTGAATACAAGTAATATGTAATAATAC
ATTATAATTTAATAACATTTAGAATAATTACATTTTATACAAAAATAAAATTA
AGAtaaaattcacatagtgcaatggtgASTAAGATGTGAAAAGACAATAAGAATAAACAGC
ATTAAAATTATTGATAGAGTTTGTAAAACCCCTAGAGATTAAGGAAAAACAAA
CATAGGAATAAATTAGAAAACTAGAGACAATAATAATTCTGTAAATTATAG
GCTACCAAAACCAGAATaagaataaacaaggactcaaaaaaac

For: 5'-3' = aaactgggacacttgtaatgaat

New R 5'-3'-taaaattcacatagtgcaatggtg

Rev 5'-3' = gttttttgagtccttgtttattctt

M103 = B9.117new (463 bp) C to T at position 259

For: 5'-3' = cagtaagtgaactcacacataattcc

Rev 5'-3' = ccagttttatttcagtttcacagc

M104 = DYS257a (288 bp) Duplicated locus. Most men have both A and G alleles at position 162, however some have only A allele. The second site at position 202 is often just C, although sometimes both C and T alleles occur.

GaacttgtcgggaggcaatGGTGACATTCATTGTGACCTTAGCCAGAGCTCACAATCAA CCATGGTGCACTGAGACTAGCTCATGCACATTCATCAGGCAGATTCAGGCAC CTGGCTGTCAGAGCTGTCAGCCTTCCTCAGTAGAGGAAAATGCTACAGTCRG CACTGGCCTGGTATCAGGAAAATAGATGCCTGCAAAAAYCCACTGTGGGACC CTAAAAGTCTTGACCTCAGGTCCCCTTTGTGCTGTCTCTGTTGTCAGGATccacta aaggaggaagtgtatca

For: 5'-3' = gaacttgtcgggaggcaat Rev 5'-3' = tgatacacttcctcctttagtgg

M105 = B9.6-7a (572 bp) C to T at position 478

For: 5'-3' = gggaggcaacctaagaaag Rev 5'-3' = aggtgaacgcattctgtcat

M106 = B9.6-7b (572 bp) A to G at position 411

For: 5'-3' = gggaggcaacctaagaaag Rev 5'-3' = aggtgaacgcattctgtcat

M107 = B9.112n (376 bp) A to G at position 298

For: 5'-3' = caaaagcactcgggttcct Rev 5'-3' = cttactcccacttatgcaacg

M108 = B9.113n (321 bp)T to C at position 40. Probably recurrent

AgatggagccagcagaaagGAGAGAAGTAGATGAACATCYGAAACTATACCTGAATG
TCAGAGAAAAGTGGATTGACTTCAGAGGAACAGCTTGATGGTGTAACTTTGG
AGAAGAATCCGGCTGGAGACTTTAGTGATCTGGGTAGAAGATAAAATCATCC
ACAATATTTACTGGGGTTTTTTTTTGCATTTCCTGAATTTGAATCTTGGCCAGAG
TAAAGGGAAATATTCATCCCTCCTCCTTTTTAGCACCCATTCCCACTTAAAGC
CACCTCTATCACATAAAATCCTCCACATTTaccatcattcaattcatctgtgt

For: 5'-3' = agatggagccagcagaaag Rev 5'-3' = acacagatgaattgaatgatggt

M109 = G3.15 (312 bp) C to T at position 264

GggtatcaaatgtcttcaacctAAAGTACAAGGAATTATTTCTCAGTGTTTTGGAATGACTT
GACTTCCTTGAAAATATTGTTGCAGAGTTGGGGACTACTTTTAAAATATCCTC
CATTGAATGTAATTCTACATGAAAGCTTGATTTTTCAAGTGCAAAATGCAAGT
GAGAAATAAGGCATATCATTCATTAAACCCTAATTCCAGCACTTTTAAATGA
GCTACTTTCTTGTATAAATATTTTAGCTATTAAGGAACAAATTGTYGCTTAAGA
AATGTATCTATCTTAAAAAATgcaagtagcaggaaattccc

For: 5'-3' = gggtatcaaatgtcttcaacct

Rev 5'-3' = gggaatttcctgctacttgc

M110 = B9.86n (389 bp) **T to C** at position 241

For: 5'-3' = cagggaaggaccgtaaaagg Rev 5'-3' = atgtttatcatgtgcagtaaaggtt

M111 = G3.19 (393 bp) -2bp (TT) deletion at position 188-189 interval. Polymorphic STS = 391 bp.

For: 5'-3' = aatcttctgcaaagggttcc

Rev 5'-3' = cagctacaaaacaaaatactggac

M112 = G3.17a (445 bp) G to A at position 286

For: 5'-3' = actttttccaacagttatttttga

Rev 5'-3' = tatatttcttgatgatgagaccaat

M113 = G3.17b (445 bp) A to G at position 112

For: 5'-3' = actttttccaacagttattttga

Rev 5'-3' = talatttcttgatgatgagaccaat

M114 = G3.23 (434 bp) T to C at position 387

For: 5'-3' = ttaccacacagttgagtagttctaaa

Rev 5'-3' = caaataaaattggagcggtta

M115 = G3.22 (413 bp) C to T at position 201

agtttacagtcacatcaatttggaAAGTCATACAAATATTGTCAAAAAAACTGATCTGAATCA
AATATGCCATGCTTGTTTCTTAATCCATTGAAGTTTTACTTATCATTTAAATGA
CTTGACAATATTAGTCAGTTTATATTTTCTTTTATGTAGATATTATGGGCTCCA
GAGTTTAAATTAGTATTTGATTTCACATTAYGAAACCATTATAAAAAAAGTCTC
AAATTAAGATAATTTAAGGTGATGAACACACAAACGTACACTTTGAAAGGAG
AAGGCAATGAAAACATGCATTCCAATAAAGGGGGAAAATGAGGCTGATGTG
CAACATAGTTGGGGAAATTGGTAAGAAGCTTTCTGTTACCACACAGTTGAGT
AGTTCTAAAAaaacagagatatggtagaaaaagga

For: 5'-3' = agtttacagtcacatcaatttgga Rev 5'-3' = teetttttetaccatatetetgttt

M116 = G3.25a (429 bp) Three alleles. A to T (M116.2) or A to C (M116.1) at position 176

TTTTCTACCCAAATATAAATAATTTGTTTTAGCCATATTATCTCATTACTGAAG TATCATAGGATGACTGAGTAGACtgctcattgtaaaatctaactgaat

For: 5'-3' = aagtatgacttatgaagtacgaagaaa

Rev 5'-3' = attcagttagattttacaatgagca

For: 5'-3' = aagtatgacttatgaagtacgaagaaa

Rev 5'-3' = attcagttagattttacaatgagca

M118 = G3.29 (478 bp) A to T at position 109

For: 5'-3' = attclaagtttcacttcctgatcc

Rev 5'-3' = tagttccctaaattacgctacctc

M119 = G3.32 (330 bp) A to C at position 224

 $\textbf{ATTT} \underline{\textbf{M}} \textbf{GCCTGTATGCTGAATTGGAATAACCCATAACATTTTCTACATCTA} \\ \textbf{ATTTAAAAAAACGGTTTAAATTTGTATTAATTaagaatacatcttgtatattgtgtgaa}$

For: 5'-3' = gaatgettatgaattteecaga Rev 5'-3': tteacacaatatacaagatgtattett

M120 = B9.87b (495 bp) T to C at position 224

GagcttggactttaggacggGGAAAAGAAGTGCTAAATGTTTTTGAATAAAACCTTTACT GCACATGATAAACATCCCTTAAAAAATTACCTAGGAGCACCCTAAATTTTAAA ATGATCACAAAGACCTGGACAGATTACAGTAAACCTTCAACATCGCTAAACA CACGTACCATAAATCAAAAGAAACACACTGCTAATGATCCGTTTTTTGATGT For: 5'-3' = gagcttggactttaggacgg Rev 5'-3': aaactttaaggcacttctggc

M121 = B9.87c (495 bp) 5 bp deletion at position interval 183-187

For: 5'-3' = gagcttggactttaggacgg Rev 5'-3' = aaactttaaggcacttctggc

M122= G3.27a (393 bp) T to C substitution at position 73

TggtaaactctacttagttgcctttTGGAAATGAATAAATCAAGGTAGAAAAGCAATTGAGA
TACTAATTCAYGCTCTCAGGGGAAAATCTGAATAAAGCTATCTTTTCTAACA
CAGAGCAAGTGACTCTCAAAGTCACAGTATCTGAACTAGCATATCAGCATCG
CCTGAATACCTAGAAATGCAAATTCCTGGGCAACACCAGAATCTAACAAAGC
AAAAAACTATGGGGGGAACAGGGAAGTCGGTTTAATAATACTGAGTTTGTGC
AACCTCAACTTTGCTTTATAGGAAAGCAAAATCTCAATATGATAAAGTTTTCT
TCAACAAAACTCTGAGATAACTATGTTGAGGGAAAGAAGTTGATCACATgcaag
aaaatctaattcgctg

For: 5'-3' = tggtaaactctacttagttgccttt Rev 5'-3' = cagcgaattagattttcttgc

M123 = G3.27b (393 bp) **G** to A at position 161

TggtaaactctacttagttgcctttTGGAAATGAATAAATCAAGGTAGAAAAGCAATTGAGA
TACTAATTCATGCTCTCAGGGGAAAATCTGAATAAAGCTATCTTTTCTAACAC
AGAGCAAGTGACTCTCAAAGTCACAGTATCTGAACTAGCATATCARCATCGC
CTGAATACCTAGAAATGCAAATTCCTGGGCAACACCAGAATCTAACAAAGCA
AAAAACTATGGGGGGAACAGGGAAGTCGGTTTAATAATACTGAGTTTGTGCA
ACCTCAACTTTGCTTTATAGGAAAGCAAAATCTCAATATGATAAAGTTTTCTT
CAACAAAACTCTGAGATAACTATGTTGAGGGAAAGAAGTTGATCACATgcaaga
aaatctaattcgctg

For: 5'-3' = tggt:aaactctacttagttgccttt Rev 5'-3' = cagcgaattagattttcttgc M124 = G3.27c (393 bp) C to T at position 246

TggtaaactctacttagttgcctttTGGAAATGAATAAATCAAGGTAGAAAAGCAATTGAGA
TACTAATTCATGCTCTCAGGGGAAAATCTGAATAAAGCTATCTTTTCTAACAC
AGAGCAAGTGACTCTCAAAGTCACAGTATCTGAACTAGCATATCAGCATCGC
CTGAATACCTAGAAATGCAAATTCCTGGGCAACACCAGAATCTAACAAAGCA
AAAAACTATGGGGGGAACAGGGAAGTYGGTTTAATAATACTGAGTTTGTGC
AACCTCAACTTTGCTTTATAGGAAAGCAAAATCTCAATATGATAAAGTTTTCT
TCAACAAAACTCTGAGATAACTATGTTGAGGGAAAGAAGTTGATCACATgcaag
aaaatctaattcgctg

For: 5'-3' = tggtaaactctacttagttgccttt

Rev 5'-3' = cagcgaattagattttcttgc

M125 = B9.108a (367 bp) T to C at position 301

Gccaccctcttatgcc1ctGGCCTTTACAAAGACAGCTGGTAAGAGGCTGCCCAGCTCAT CTGAAGTACAGGATAAGATTGTCTGACTTGGAGATACCATTTTCCACTTAGCA GCCATGTAATCTTTCATATTCATTTTTTCTAAGTGGCACTTTTCTCAGATGTAA AATGGGGATAATGAGTTTATTCATCTTTGAGTTGCTCCCAAGCAGAAGTCAAC TTGAGACTATAAACTTGTGCTCACTGCAGTGCTTGAAACCGAGTTTGTACTTA ATAAATAGCTGCATACATCTTTTTCTAYACATGTCAGATGCTTAATTGTGTTT CCCGAAGATGTTGCCAAGCCgggtcctcacataactcctga

For: 5'-3' = gccaccctcttatgcctct

Rev 5'-3' = tcaggagttatgtgaggaccc

M126 = B9.108b (367 bp nominal) **4 bp deletion** (AATA) at interval 277-280.

GccaccctcttatgcctctGGCCTTTACAAAGACAGCTGGTAAGAGGCTGCCCAGCTCAT CTGAAGTACAGGATAAGATTGTCTGACTTGGAGATACCATTTTCCACTTAGCA GCCATGTAATCTTTCATATTCATTTTTTCTAAGTGGCACTTTTCTCAGATGTAA AATGGGGATAATGAGTTTATTCATCTTTGAGTTGCTCCCAAGCAGAAGTCAAC TTGAGACTATAAACTTGTGCTCACTGCAGTGCTTGAAACCGAGTTTGTACTTA ATA**AATA**GCTGCATACATCTTTTTCTATACATGTCAGATGCTTAATTGTGTTT CCCGAAGATGTTGCCAAGCCgggtcctcacataactcctga

For: 5'-3' = gccaccctcttatgcctct

Rev 5'-3' = tcaggagttatgtgaggaccc

M127 = G3.30 (412 bp) C to A at position 372 bp

TgaaaggaaatcagtgtaagagcTAGAGGTAGCGTAATTTAGGGAACTAATCAGGAAAGA GGTATTAACATTTCTGAATCCTTAGTTTCACTTATCCTTTCAATTCACAAGATT GCTTTATTTCACATTTTGATAAAGACCAAAATGGTCCAAAAATAAGGGGAGG AAGAACCTATACTACAAGAACCGAATTCCCAGACACTCAGGATAAACTTTAG GTATATCCTTCAATCAGCTTTGTTCCAAATACAGGTAACGAGCCAGGCAATGT TACGGAAAATAAGGGTAAGATAAAGCAAATATCCTGTGCTTTGGTTAACAAA CAAAACTGTATCACAAGTCAAACTCGTACAAAAGGCAGGAGAAGAGGT**M**TG GAAGATCTGTTAGGtgctgaactacagtcacctttaca

For: 5'-3' = tgaaaggaaatcagtgtaagagc

Rev 5'-3' = tgtaaaggtgactgtagttcagca

For: 5'-3' = actttttccaacagttattttga Rev 5'-3' = tatatttcttgatgatgatgagaccaat

M129 = A8.04 (255 bp) **G** to **A** at position 221.

For: 5'-3' = aatggcttactacaaagaacatttc Rev 5'-3' = tacacggtctctaccaaagaaga

M131 = A8.14n (306 bp) 9 bp deletion at interval 93 to 101

CacacccagaatacaataattttAAAAACATAATAAAGGTCAATTTAGAGCAGAGAAATTA
TTCTTTTAAATTACAAATGTTTGCTGTTCAG**GCAAATTAC**ACAGAAAGGTTA
AGAATAACCCTTTAAATGATAGGAAAAGGCATTAGTAAGATAAAATGTGATT
ACTATTGAGATAAATATTTGCTATAAAAAATAATTCAATTTGGTTAAACACAAA
TTGACTTCTTAAATAATCTTAAACATTAAGTAGAAGTAATTTTAGCTTATCAG
TAAATTTGAgaaaatgtacacttgtagaataaaaag

For: 5'-3' = cacaccagaatacaataatttt Rev 5'-3' = ctttttattctacaagtgtacattttc

M132 = B9.67b (568 bp) G to T at position 482

For: 5'-3' = aacagaattatcaggaaaaggttt Rev 5'-3' = ttttacttgttcgtgtactttcaa

For: 5'-3' = tgaaatggaaatcaataaactcagt Rev 5'-3' = cetttetttttetttaaccette

For: 5'-3' = agaatcatcaaacccagaagg Rev 5'-3' =tetttggettetetttgaacag

M135 = A8.08F-newR (211 bp nominal vs 212) 1 bp insertion (+ C) at position 150 = site c, within homopolymer A track.

Site a (A)₁₀ -TTT most males

Site c (A)9CATTT = M135

Site d (A)11TTT

For: 5'-3' = tgaaatggaaatcaataaactcagt

Rev 5'-3' = ccttttctttttctttaacccttc

M136 = B9.61 (339 bp) C to T at position 196

AtgtgaagacaacactgtgtggGAGAACCTAGGAAAGTAATTTTACATGCTAAAATGAGT
TTCCCTAGTTAATGTTAACATGAACTACCAACCGTATTACCTTCTCCTCAGGA
GATAAGTTTTGTTTGCTATTGCTGACAGGAAAGCCACTGCCAAATTCTTTGGA
ATGAATATCAGCTCCATATTCAACTGTCAYGTCTTCCTCAATGCTGCTCACCA
GCCTCCAGAATTCCTTCTCTACAAGTTCTGTAGGCACCATCTGTGAAAACACA
TGTAAAAGGTTATCATAGCCCACTATACTTTGGACTCATGTCTccatgagaactaagac
taccacaa

For: 5'-3' = atgtgaagacaacactgtgtgg Rev 5'-3' = ttgtggtagtcttagttctcatgg

M137 = G3.27d (393 bp) **T to C** at position 289

TggtaaactctacttagttgcctttTGGAAATGAATAAATCAAGGTAGAAAAGCAATTGAGA
TACTAATTCATGCTCTCAGGGGAAAATCTGAATAAAGCTATCTTTTCTAACAC
AGAGCAAGTGACTCTCAAAGTCACAGTATCTGAACTAGCATATCAGCATCGC
CTGAATACCTAGAAATGCAAATTCCTGGGCAACACCAGAATCTAACAAAGCA
AAAAACTATGGGGGGAACAGGGAAGTCGGTTTAATAATACTGAGTTTGTGCA
ACCTCAACTTTGCTTTAYAGGAAAGCAAAATCTCAATATGATAAAGTTTTCTT
CAACAAAACTCTGAGATAACTATGTTGAGGGAAAGAAGTTGATCACATgcaaga
aaatctaattcgctg

For: 5'-3' = tggtaaactctacttagttgccttt Rev 5'-3' = cagcgaattagattttcttgc

M138 = A8.17(442 bp) C to T at position 291

AacttccaaaactgtgaaaagattGTTTTTAAAAGGCTATAACAGTGACTTTCAGGTGAAGA CTTGGACAAATAGATAATTTCTGTACCCATTAAAATCAGGGGCTGTTACTATG TTTGAAGACATTGTCGCCACAGCTTGAAGTCTGTAAGGAAAACCTGTAAAAT TAGTGGGTGCCCACTCTAGTTTTAATCATTTGAGTTTCCACTCCTCATTGTGGT TGAACTATTTTATAACTCTGCAAAATCTAGAAAGTTGAAAAGAAACCAAAGA TACTTTCCCTTTTCTTCYCACTTCCTACCCTTGGCCCACCTCCTTCTCCACC TACTACTCCACATGGAACCTGGAGATTTGAGTCGGGGAGTGATGTAATACCT GCGGCGCGTTGGCCCTTTACACACCTGTCAGCCATTTCAAGGCctgaaggggctgcttt aatc

For: 5'-3' = aacttccaaaactgtgaaaagatt Rev: 5'-3' = gattaaagcagcccttcag

M139 = A8.28a (459 bp nominal vs 460) 1 bp deletion at position 401. 5 G's to 4 G's.

For: 5'-3' = ttactgataatgccatattgttttg Rev 5'-3' = ttccagacaccaatggtcct

M140 = A8.28b (459 bp nominal vs 460) 1 bp insertion within 9 A's homopolymer (most men) to 11 A's at position 73. Recurrent because 11 A's found in different haplogroups.

For: 5'-3' = ttactgataatgccatattgttttg

Rev 5'-3' = ttctcagacaccaatggtcct

M141 = A8.30a (424 bp nominal) T to A at position 51. Locus also has two homopolymer T tracks which are both polymorphic. See next below.

For: 5'-3' = catcttaaaatacatttcatagcttt

Rev 5'-3' = gcttactattaggtctagcatcct

For: 5'-3' = catcutaaaatacatttcatagcttt (

Rev 5'-3' = gcttactattaggtctagcatcct

M143 = B9.50b (385 bp) G to T at position 246

For: 5'-3' = atgctataataactaggtgttgaag

Rev 5'-3' = aattcagcttttaccacttctgaa

M144 = B9.99 (452 bp) T to C at position 342

For: 5'-3' = agcacaagggtcacattgag

Rev 5'-3' = aggacaaggctttttgttgtt

M145 = A8.05b (208 bp) G to A at position 166

RAGAGCCAGCCTTAGCCTAATCaagaaccatgatccaaaaagg For: 5'-3' = ttcagcaagagtaagcaagagg

Rev 5'-3' = cctttttg.gatcatggttctt

For: 5'-3' = gaatggggtgttacatggaga

Rev 5'-3' = cctattatgagagacctgttttcc

M147 = G3.35 (439 bp nominal) 1 bp insertion (extra T). Associated with GTTT repeat. 3 T's to 4 T's at position 116. Locus also has T homopolymer which cause stutter bands during PCR.

For: 5'-3' = gtattctggggcaattttagg Rev 5'-3' = ttgatacaagaggttattttaagca M147new = G3.35 (276 bp nominal) 1 bp insertion (extra T). Associated with GTTT repeat. 3 T's to 4 T's at position 97.

M148 = B9.67c (568 bp) A to G at position 314

For: 5'-3' = aacagaattatcaggaaaaggttt

Rev 5'-3' = ttttacttgttcgtgtactttcaa

M149 = B9.67d (568 bp) G to A at position 469

For: 5'-3' = aacagaattatcaggaaaaggttt

Rev 5'-3' = ttttacttgttcgtgtactttcaa

M150 = B9.18 (289 bp) C to T at position 146

For: 5'-3' = gcagtggagatgaagtgagac

Rev 5'-3' = cctactttcccctcttctg

M151 = B9.58b (422bp) G to A at position 209.

ActtaatttatagtttcaatccctcaGTAATTTTAACTTACTTCTATTTTAAGAACTATAACCA AACTATCTGTAAGACTTTTAAGCACTATCATACTCAGCTACACACATCTCTTAAC AAAAGAGGTAAATTTTGTCCTTTTTTGAACGTCATAGAGTATACTCACACAAA CCAAGAAGAAACAATCTACTACATACCTACGCTATATGRTATATAACTATTG CTCCTAGGCTACAAATTAGTGCGACACTATTGTACTGAATATTATAGGCCATG TAACACAATGGTTTAAGTATCTGTGCCTCTAAACACAGAAAAGATATAGTGA AAGTACAGTATTGCTCCTTTATTAAACTCAAAATGTTATGCAGCATATGACCG ACTATAAAATAGCGCTTATCcagatacagacatctccatgaa

For: 5'-3' = acttaatttatagtttcaatccctca

Rev 5'-3' = ttcatggagatgtctgtatctgg

M152 = B9.13 (287 bp) C to T at position 101

AagctattttggtttcctttcaAGAAAGGGCTGTGGTCTGTGGAAGGTGTCAGGAACATATT
TTCCACGGTCTGCTTTCTCCTGATAATGTTCTTCTTYGGCCCACCTGAGAC
ATAATCCCTGAGCTCCGAGCCCTTTTTGACTGAAGCTCCTGTTGAACAAGATT
CTCAACGTTTCTACCCTGATCCACCTTCTGCCGCCGCCGTCGCCTCTCCAGAG
CCCGGCTCCTTGTCCGACTCCCTTGATGTTCAAATTTTTCCAGCTGcaatcatacccac
acaaggc

For: 5'-3' = aagctattttggtttcctttca

Rev 5'-3' = gccttgtgtgggtatgattg

M153 = A8.28c (459 bp nominal) T to A at position 427 bp

For: 5'-3' = ttactgataatgccatattgttttg

Rev 5'-3' =ttctcagacaccaatggtcct

M154 = B9.58c (422bp) T to C at position 252.

ActtaatttatagtttcaatccctcaGTAATTTTAACTTACTTCTATTTTAAGAACTATAACCA
AACTATCTGTAAGACTTTTAAGCACTATCATACTCAGCTACACACTCTCTTAAC
AAAAGAGGTAAATTTTGTCCTTTTTTGAACGTCATAGAGTATACTCACACAAA
CCAAGAAGAAACAATCTACTACATACCTACGCTATATGGTATATAACTATTG
CTCCTAGGCTACAAATTAGTGCGACACTAYTGTACTGAATATTATAGGCCAT
GTAACACAATGGTTTAAGTATCTGTGCCTCTAAACACAGAAAAGATATAGTG
AAAGTACAGTATTGCTCCTTTATTAAACTCAAAATGTTATGCAGCATATGACC
GACTATAAAATAGCGCTTATccagatacagacatctccatgaa

For: 5'-3' = acttaatttatagtttcaatccctca Rev 5'-3' = ttcatggagatgtctgtatctgg

M155 = G10.57c (327 bp) G to A at position 251

For: 5'-3' = tctctaacttctgtgagccac Rev 5'-3' = ggaaaaactaaactctaaatctct

M156 = A8.05c (208 bp) A to G at position 147. Linked to M145 derived allele. TtcagcaagagtaagcaagaggCACTGAGCCGCTGGAGTCTGCACATTGATAAATTTACT TACAGTCGTAAATAAATTGCATCATCTTCAGCTAGTAACACAGAGTCTAATTT TTATAGCGGCATACTTGCCTCCACGACTTTCCTRGACACCAGAAAGAAAGGC GAGAGCCAGCCTTAGCCTAATCaagaaccatgatccaaaaagg

For: 5'-3' = ttcagcaagagtaagcaagagg

Rev 5'-3' = cctttttggatcatggttctt

M157 = B9.12b (352 bp) A to C at position 176

For: 5'-3' = getggeaagacaettetga Rev 5'-3' = aatatgtteetgacaeettee

For: 5'-3' = tgaaatggaaatcaataaactcagt

Rev: 5'-3' = ccttttctttttctttaacccttc

M159 = G10. 83new b (190 bp) A to C at position 89

For: 5'-3' = attggattgatttcagccttc Rev 5'-3' = attttattttctgtgttccttgc

M160 = B9.47b (361 bp) A to C at position 251

CagaataataggagaatttttggtCAAATAAAAGGCCATATTATATTTCTTTTGATAAAAGT ATCATGTGTTCAGTATGTTTTATTATTTGAAATAATTAACATGACAGGAATAT ATTTGAAAAAAATTCCAAAAAAAAGCTAAATATACAAACTAAGAAAAATTATAT GATTATACTTATCTGCAGTATTGTAAAACAATAGTTCCAAAAACTTCTGAATT ACAAGTTTAATACATACAACTTCAATTTTC**M**ACTACATTGTGGTTAGACGTT CAGAGGAATCACAAAGGACCTCAACATGCTAGATAAGAAAATGTATTTTTA AATGTTTTGGCTCAgctgcttagaaaataaggaaaat

For: 5'-3' = cagaataataggagaatttttggt Rev 5'-3' = attttccttattttctaagcagc

M161 = A8.05d original (460 bp) C to A at position 111

For: 5'-3' = tcacagcagcttcagcaaa

Rev: 5'-3' = cctttttggatcatggttctt

new R 5'ataaaaattagactctgtgttactagc3'(used with F primer, just amplifies the first 2 sites including homopolymer T region.

M162 = DYS257b (288 bp) =

C/T at position 202), most men are just C at position 202

Duplicated locus. Most men have both A and G alleles at position 162, however some have only the A allele. The second site at position 202 is often just C, although sometimes both C and T alleles occur on a chromosome background that is both A and G at position 162.

GaacttgtcgggaggcaatGGTGACATTCATTGTGACCTTAGCCAGAGCTCACAATCAA CCATGGTGCACTGAGACTAGCTCATGCACATTCATCAGGCAGATTCAGGCAC CTGGCTGTCAGAGCTGTCAGCCTTCCTCAGTAGAGGAAAATGCTACAGTCRG CACTGGCCTGGTATCAGGAAAATAGATGCCTGCAAAAAYCCACTGTGGGACC CTAAAAGTCTTGACCTCAGGTCCCCTTTGTGCTGTCTCTGTTGTCAGGATccacta aaggaggaagtgtatca

For: 5'-3' = gaacttgtcgggaggcaat Rev 5'-3' = tgatacacttcctcctttagtgg

M163 (340 bp) G10.35b A to C substitution at position 168

GcagcatataaaactttcaggACCCTGAAATACAGAACTGCAAAGAAACGGCCTAAGAT
GGTTGAATCCTCTTTATTTTTCTTTAATTTAGACATGTTCAAACGTTCAATGTC
TTACATACTTAGTTATGTAAGTAAGGTAGCGCTTACTTCATTATGCATTTCAA
TMCTCAAAAAAAAATTCCTTTGTGAAATGTTGAAATATTTTTCTAATCTGTTTC
ACGAGCTTCAAAAAATGAGGAAAAAAGATTCAGTTTACATTTCAGCAAAATGC
CTCTTTTTAATCGGATTTATGTTTACTTAACATTTACAGTACATTTACgcttgagcaa
agttaggtttt

For: 5'-3' = gcagcatataaaactttcagg Rev 5'-3' = aaaacctaactttgctcaagc

M164 = G10.100b (493 bp) T to C at position 329

TagaagtagcagattgggagaggACATGTGTTCAAGTTGTACTACTTGTATGTCTTGTTTA
GATATTACAGTCTTTTTCTTTTATCAGAAAAATAATTGAATAATGATAAAATCA
GTTGCAGATTAAGACAGATTATCTGTTGCAGTCTTCTCAAAACTTAATTTAAG
TACATTATTTTCAGCTAGCATTTCTTCCTTCACATAGAACCTCCATGTGTGGA
GGGATTTCCTAATGAGTCTATTGTATGTACAATAGCACTTAATGACATAGCTT
TTAAATAATAACAGGATTTTACCAAATGTTTAATATGTGCCAGGCATCAAGC
ACCYTACACAGTTTAATTATTGCATAGATTTGGACAGCAACTCTGCAAGTTA
GGTATGGTCATGAACCTTTGCAGATAAGGAAACTGTGTTTCACAAGGAAAG
AAATTGTCCTGGATCATACAATAAGCTAGGATTTGCTCCAgaccattttttcattttatcagg

For: 5'-3' = tagaagtagcagattgggagagg Rev 5'-3' = cctgataaaatgaaaaaatggtc

M165 = B9.008c. (340 bp) A to G at position 132.

AaagcgagagattcaatccagGATGACAGAATGCGTTCACCTTTAAAGGGATTAAAAGA AGTATAATACAGTCTGTATTATTAGATCACCCAGAGACACACAAAACAAGAA CCGTSAATTGAATTAGTGGTATACTAATAGAGTGGTTTTACCTGAAATATTTA CACATCAATCCTACTGAATTCTTACAACAAATGATTTAGATTAGCTATTGTAT TCACCAGTTGAAAGAACAGAAAATATTGAGGGAGATAACTTGTGTCAGTGCA ACTTAATCAGATTTAGGACACAAAAGCAACTACATAATGAAAAAAGAGAgctggt gacttaacttgctaaaa

For: 5'-3' = aaagcgagagattcaatccag Rev 5'-3' = ttttagcaagttaagtcaccagc

M166 = G3.27e (393 bp) G to A at position 53

tggtaaactctacttagttgcctttTGGAAATGAATAAATCAAGGTAGAAAARCAATTGAGA
TACTAATTCATGCTCTCAGGGGAAAATCTGAATAAAGCTATCTTTTCTAACAC
AGAGCAAGTGACTCTCAAAGTCACAGTATCTGAACTAGCATATCAGCATCGC
CTGAATACCTAGAAATGCAAATTCCTGGGCAACACCAGAATCTAACAAAGCA
AAAAACTATGGGGGGAACAGGGAAGTCGGTTTAATAATACTGAGTTTGTGCA
ACCTCAACTTTGCTTTATAGGAAAGCAAAATCTCAATATGATAAAGTTTTCTT
CAACAAAACTCTGAGATAACTATGTTGAGGGAAAGAAGTTGATCACATgcaaga
aaatctaattcgctg

For: 5'-3' = tggtaaactctacttagttgccttt Rev 5'-3' = cagcgaattagattttcttgc M168 = DFFRY Ex01B site a(473 bp) C to T at position 371 noncoding

For: 5'-3' = agtttgaggtagaatactgtttgct Rev: 5'-3'= aatctcataggtctctgactgttc

M169 = DFFRY Ex01B siteb (473 bp) T to C at position 97 noncoding

For: 5'-3' = agtttgaggtagaatactgtttgct Rev: 5'-3'= ccagggcccgagggactctt

M170 = DFFRY Exon08 (405 bp) A to C at position 327

For: 5'-3' = tgctlcacacaaatgcgttt

Rev 5'-3' = ccaattactttcaacatttaagacc-3'

M171 = DFFRY Ex01B sitec (473 bp) G to C at position 440 noncoding?

TGAAGAATTAAAACAATAGTTTTAGCAGTTTGGGTAAGAGATGTTTACAGAA ATGTTTTGTG**S**AATAAAACtgaacagtcagagacctatgagatt

For: 5'-3' = agtttgaggtagaatactgtttgct Rev: 5'-3'= ccagggcccgagggactctt

M172 = DFFRY Ex45 (345 bp) T to G at position 197

TtgaagttacttttataatctaatgcttAATCTCTTTAAATATTTAAAATTAGGAGCCAGATGACCAGGATGCCCCAGATGAGCATGAGCCCTCTCCATCAGAAGATGCCCCATTATATCCTCATTCACCTGCCTCTCAGTATCAACAGGTAAAAAGGATTTTTCATTTTATCCCCCAAACCCATTTTGATGCTT**K**ACTTAAAAGGTCTTCAATTATTTTCTTAAATATTTTGAAAGTCCAAACTTTCTCTGTACCTGGCTGATATTTAAAACTGGATAAACTGTTCCAAACCAACATGGAGTGAAGATGGATccactgtgactgtaaagtaataaattat

For: 5'-3' = ttgaagttacttttataatctaatgctt Rev: 5'-3' = ataatttattactttacagtcacagtgg

For: 5'-3' = aagaaatgttgaactgaaagttgat

Rev: 5'-3'= aggtgtatctggcatccgtta

M174 = DffryEx38 (348 bp) **T to C** at position 219

AcateteagategttgtttggtTCATAAAAATCTGTTTCTTCCATGTACCAAGCAAAATAAA CACATCACTAAAATTTGACGTTCATAGATGTTTCTGTTTTAGGTATGATGCAC TGTGCGTTCTTCCGTCACAGCAAAAATGTACGTTTTTGGTTTACTCATAAT GTCCTTTTTAATGTATCAAATCGCTTCTCTGAATACCTTCTGGAGTGCCCYAG TGCAGAAGTGAGGGGTGCATTTGCAAAACTTATAGTGTTTATTGCACACTTTT CCTTGCAAGATGGGTCTTGTCCTTCTCCTTTTTCCATCAGGACCTTCTAGTc aggtaattgcatggcttttt

For: 5'-3' = acatctcagatcgttgtttggt Rev: 5'-3'= aaaaagccatgcaattacctg

TGCTATACACCAATGCACTACACTCGACCAAATAAAATTACTGTAATTCCAA ATTTATTTGAAAATGTAAGTGCTAATCAAGTTATTtccctgagatagttaagaatggag

For: 5'-3' = ttgagcaagaaaaatagtaccca

Rev: 5'-3'= ctccattcttaactatctcaggga

M178 = G10.72b (514 bp) C to T at position 220

For: 5'-3' = taagcctaaagagcagtcagag

Rev 5'-3' = cagagggagcaatgaggaca

M179 = Dffry exon 07 (426 bp) C to T at position 316

For: 5'-3' = acactactgtgctgtaatttgtgaa

Rev 5'-3' = agacaggttcacatcactaatgg

M180 = Dffry exon 11(447 bp) T to C at position 402

For: 5'-3' = acactactgtgctgtaatttgtgaa

Rev 5'-3' = tggtaagatttctctacatgaacag

M180 = Dffry exon 11(232 bp) T to C at position 128

CaggacettgataatatetgGGCAGCACAGGTAAGAAAGTGAGATGATAGCTATTTTCTA AGAAAGATACCAAAAAGGAGAAAATTTTTGGTAACCCTTATATAATGGCCAG

CAATTTAGTATTGCCYGACTTTTACTAATGCATGTGctgttcatgtagagaaatcttaccaAGAATTTTTAAACAAAAAAAAAAAATTTTTCTGTCTTTgtatatatattcatggtagcaa

NEW F 5'-3' =caggaccttgataatatctg

NEW Rev 5'-3' == ttgctaccatgaatatatatac

M181 = Dffry exon 12 (294 bp) T to C at position 130

GcttttatttattctacttttgtttttTCAACAGGCAGGAAAACATGAAGCCATTGTGAAGAATG
TACATGATCTGCTAGCAAAGTTGGCTTGGGATTTTTCTCCTGGACAACTTGAT
CATCTTTTTGAYTGCTTAAGGTAGTAGCTTGAATAGTAAAGTATTGCCAAAT
AGTAAATATTGCCAGTTAATTCTAAGTAAAGTTTAATTCGTTAGATTTCTTTT
GCTTATAGCTAGTGTGCTTAACTAACATTTTCATGGAAGAATCTCTGatgaaaaaga
attggtcattgtt

For: 5'-3' = gettitatttattetaettttgttttt

Rev 5'-3' = aacaatgaccaattctttttcat

M182 = Dffry exon 13 (364 bp) C to T at position 38

TattcaaagacttaaagcagtggttaATGTAAACAAAYGTAATAAATTATGTGGTATTTATA
TCATTTAAATACTTTCTTTAGGCAAGTTGGACAAATGCAAGTAAAAAGCAAC
GTGAAAAGCTCCTTGAGTTGATACGCCGTCTTGCAGAAGATGATAAAGATGG
TGTGATGGCACACAAAGTGTTGAACCTTCTTTGGAACCTGGCTCAGAGTGAT
GATGTGCCTGTAGACATCATGGACCTTGCTCTTAGTGCCCACATAAAAATACT
AGATTATAGTTGTTCCCAGGTATGGGAGTGTTTCTTTGTTCAGTTTTCTGACTT
TCCTTCACAAGTaggataacttagttacaagatgattcc

For: 5'-3' = tattcaaagacttaaagcagtggtta

Rev 5'-3' = ggaatcatcttgtaactaagttatcct

M183 = Dffry exon 19 (427 bp) A to C at position 324

For: 5'-3' = actgggtaaatatgactatgattgag

Rev 5'-3' = ttccttttaacctattattactttcc

M184 = Dffry exon 23 (305 bp) G to A at position 62

For: 5'-3' = cactttattttagtctgtgtctttttc

Rev 5'-3' = aaacttagtaacatctatttctcctct

M185 = Dffry exon 27 (430 bp) C to T at position 89

For: 5'-3' = ggagtacctatcactgaatgtgc

Rev 5'-3' = gtcattcatttctgcttggaac

M186 = Dffry exon 30 site a (365 bp nominal) -1 bp deletion (4G's to 3 G's) at position 62 (364 bp = mutant) 325 bp w/out homopolymer

For: 5'-3' = ttgcatttactgttctagagagttct

Rev 5'-3' = ccacagagcaagactccatc

newRev 5'-3'=gtacttactttaatgagattagcac Homopolymer clipped off

M187 = Dffry exon 30 site b (366) IGNORE Homopolymer in tree T(10 to 11 T's) 325 bp w/out homopolymer

TigcatttactgttctagagagttctCAAAAAGAAATAGGAAACCACTTGAACAGTTTGGGGA AGTTGTATAGAAGATCTCATTTCCTTCCAGCTCTCTGTTCTCCTAACTCCTTGT CCTTTTCTATCTCCATGTTGTGAGTTGGGCCTATAATATTTTTCCTTTTGCAGG ATAATGTTAAAAACACAGGTGAAACAGGTGTCGAAGAGCCAATACTGGAAG GCCACCTTGGGGTAACAAAAGAGTTATTGGCCTTTCAAACTTCTGAGAAAAA GTATCACTTTGGTTGTGAAAAAAGGAGgtgctaatctcattaaagtaagtacTTTTTTTTTCT TTTTTTGAgatggagtcttgctctgtgg

For: 5'-3' = ttgcatttactgttctagagagttct

Rev 5'-3' = ccacagagcaagactccatc

newRev 5'-3'=gtacttactttaatgagattagcac Homopolymer clipped off

M188 = Dffry exon 31 (401 bp) C to T at position 185

GtattcctttgaagaaacatattgTTCCTAACCTATATTTTCTACTAATAACATGTAATGTCT
TTTTCTAACTTACTAGGAATTAATTGATGATTTCATCTTTCCCGCATCCAAAGT
TTACCTGCAGTATTTAAGAAGTGGAGAACTACCAGCTGAGCAGGCTATTCCA
GTCTGTAGTTCACCYGTTACCATCAATGCCGGTTTTGAGCTACTTGTAGCATT
AGCTATTGGCTGTGTGAGGAATCTCAAACAGATAGTAGACTGTTTGACTGAA

ATGTATTACATGGGCACAGCAATTACTAGTGAGTATTTTAAATTATAAAGCTGTTTTGTTCATTAATAATACTTCACTGTAAAAATTTTATTTGGTGTTTTAgaaaaaatta acttgtgatggactt

For: 5'-3' = gtattccctttgaagaaacatattg

Rev 5'-3' = aagtccatcacaagttaattttttc

M189 = Dffry exon 34 (378 bp) G to T at position 191

For: 5'-3' = actologettatgtttgtcattg

Rev 5'-3' = gettttggtacactetgetttt

M190 = Dffry exon 44 (346 bp) A to G at position 73

CtctgtcacaagtaaggaaatgatCGTGAAATTTTTGTATTAGCATTTTAAGCTGATACTGA
AAATCATTCTRAATTCTAAATAGTTTTATTTTTTTTTTCTAAAGGGTAACGGAGAT
CTTAAAAGAAAATGGACCTGGGCAGTGGAATGGCTAGGAGATGAACTTGAA
AGAAGACCATATACTGGCAATCCTCAGTATAGTTACAACAATTGGTCTCCTCC
AGTACAAAGCAATGAAACAGCAAATGGTTATTTCTTAGAAAGATCACATAGT
GCTAGGATGACACTTGCAAAAGCTTGTGAACTCTGTCCAGAAGAGGTAAAAA
AAaaaaaaggctaccaatggacag

For: 5'-3' = ctctgtcacaagtaaggaaatgat

Rev 5'-3' = ctgtccattggtagccttttt

M191 = DBY exon 2 (429 bp) T to G at position 342. Non-coding (cDNA bp# 175+120) TtgcatttgtcatggttggtTGACCTGGACATCTTTAAAATTTGGCAGGTAATACCAGGCC GACATGGCAGCTAAGTTTGTGGTACAGGATAAGATTGGAATCTAGGTCTCAT TTGTCTTTTGTGATGTTATCTGTTCTTGTGTATCAGCATGTGAGCTATTGATAT CTCTTCTAGCTTGCTAATCTGGACCTGAACTCTGAAAAACAGAGTGGAGGAG CAAGTACAGCGAGCAGTAAGTAAAAACTTTTTTTTAAAAAATGGAGTGTTTATCA GAGCTTAATGTTAATGTCTTACTGGACTTGTTAATTTTAAATTTACATTTTTT CTTTACAACTTGACTAKATGAAAAATATGAGATATTTTGGTGTGTCTGGGTAAT AAAATACACTGTTTACCTATGTCTGCTgaaaatacaaaaaattatcctggc

For: 5'-3' = ttgcatttgtcatggttggt

Rev: 5'-3'= gccaggataattttttgtattttc

M192 = DBY STS 02 (457 bp) C to T at position 202.

For: 5'-3' = catgggctgctgacatttt Rev: 5'-3'= aaatccttttggtttgtttgttt

For: 5'-3' = gcctggatgaggaagtgag Rev: 5'-3'= gccttctccatttttgacct

M194 = DBY STS 03b (426 bp nominal) T to C at position 101.

For: 5'-3' = gcctggatgaggaagtgag Rev: 5'-3'= gccttctccatttttgacct

For: 5'-3' = ccactcagctttcctcaggt Rev: 5'-3'= cgttcgtttagtcataagatcg

GGACTACGTAATCCCGcgatcttatgactaaacgaacg

M196 = DBY STS 07 (445 bp) C to G at position 330.

For: 5'-3' = ttagacaacttactactttgatgtcct Rev: 5'-3'= taaacattacatgagaaattgctgt

For: 5'-3' = tcagacagtttagttggttacttcc Rev: 5'-3'= aagacacatcctcagctaatcttt

M198 = DBY STS 08a (444 bp) C to T at position 45

For: 5'-3' = tgaggtggaatgtatcagtatacc Rev: 5'-3'= tgatttcaaggatttgttagtctt

M199 = DBY STS 08b (444 bp nominal) + 1 bp insertion (extra G) at position 404 (445 bp with mutation).

 $\label{eq:gcagcag} GCAGCAGGCTTTAATTTAATGTAGATTCATACTGCTCTGTTAAAGCTGCATTGAAATGTTAAAATGGCTTACACTTG{\color{red}{\textbf{G}}{\textbf{C}}{\textbf{C}}{\textbf{C}}{\textbf{C}}{\textbf{C}}{\textbf{C}}{\textbf{C}}{\textbf{A}}{\textbf{A}}{\textbf{A}}{\textbf{T}}{\textbf{C}}{\textbf{T}}{\textbf{T}}{\textbf{a}}{\textbf{a}}{\textbf{a}}{\textbf{c}}{\textbf{c}}{\textbf{a}}{\textbf{c}}{\textbf{a}}{\textbf{c}}{\textbf{a}}{\textbf{c}}{\textbf{a}}{\textbf{c}}{\textbf{c}}{\textbf{c}}{\textbf{c}}{\textbf{c}}{\textbf{C}}{\textbf{$

For: 5'-3' = tgaggtggaatgtatcagtatacc Rev: 5'-3'= tgatttcaaggatttgttagtctt

M200 = DBY STS 09a (429 bp) **G** to **A** at position 318

For: 5'-3' = ggcttacacttgcagactttg

Rev: 5'-3'= ggagaaatgtacaagagtctaaacc

M201 (326 bp) DBY exon 11&12 G to T at position 136

TatgcatttgttgagtatatgtcAAATTGTGACACTGCAATAGTTACTACTTGAGTTACTATA
TTAGTGCAATTAATTACACAACTATATATAGTAAttagtttctcagatctaataatccagTATC
AACTGAGGKTTTTCGTAATAGGTACTTAGTGTTGGATGAAGCTGATAGGATG
CTGGATATGGGATTTGAACCTCAGATACGTCGTATAGTTGAACAAGATACTA
TGCCACCAAAGGGCGTTCGTCACACCATGATGTTTAGTGCTACTTTTCCTAAG
GAAATACAGGTACTGTTTGAcgtttgaactttcattcagaac

For: 5'-3' = ttagtttctcagatctaataatccagt

Rev: 5'-3'= gttctgaatgaaagttcaaacg

M202 = DBYexon 16 (392 bp) T to G at position 259. Non-coding (cDNA bp# 1974+38)

For: 5'-3' = ggaattgcagggtttaagc Rev: 5'-3'= gccacccacctgtaatcc

M203 = UTY1 exon01 (1014) (503 bp) G to C at position 248; synonymous substitution, SER

CTCTCGCGGCTCGCTTTTCCTTCCGCCATTTTCTTTGCCTCATCACCGAAGGCA ACAGCGGCGTAGTGAGCGACACTGCGCASGATTTCATGGAAACAACAAATT TCCAAGTCCCACGACGATACCCAACCTTAATCGAGTAGTTGAAAAGACGCCT TCAATCGCTGCTTGAGACTGTGACGCCAATTTTATCGCCTCCTCAGCGGCTGC AAGGAAAAAAGCTGAGGCAAAGACTTAAGCTACCGAAGCACGGGCAGCGGA ACTCGGCTACCTGGATCACATCTGGGAAACTACAGGGAAGGCAGAAGCTCGC AGTGCtggagagcacagcagaattt

For: 5'-3' = gagtgccaagctgaggatga Rev 5'-3': aaattctgctgtgctctcca

New Rev 5'-3':tccttggcagccgctgaggag

M204 = UTY1 ex 02 = Intron 1 (1158-4) (286 bp) T to G at position 234; non coding AaggggcgaagtattccagAGTACGGGGACAGCAAAGGCAAAGAAACACTTTTCCGACC CCTTGGCCATGGAGCAGAGCCAAAATAAATACTGGCTGGGCGGTAAGGAAC GCGGGGCCTTGGTAGAGCAAAGTGCGGACCAAAGACTTTGCGTCTGGTTGCT TTTACCTTGCCTAGTAGGGTCTTCGTTCTGGCGCCATCTTCATGAAGCCTCAC GAACCCGAAGAGACGGCTGKAGAGAGAGAGAGACACAGAGCTTGTTAATGGTC TGAGAAAGCCAGTGACTTGCTCCTTCCCGAGTCCAAGAGCGACAGCGACAGA TTGGTGAGTGCCAAGCTGAGGATGACCCCGTCATCAACGTGGGCAAGCTGCG TCCAGGCCTTCCCGGAGAGTATCGCCAGCCAACCAGGCGGTGATGGAGGTG CGTACCTGTCCATGCCACCAAGCGCCCTCCCTTTCCTCGACTGTcaggctaacagactcct cttca

For: 5'-3' = aaggggcgaagtattccag Rev 5'-3': tgaagaggagtctgttagcctg

M205 = UTY Intron 2a (1221+3624) (541 bp) T to A at position 78.

For: 5'-3' = gtataatactgtggttggaaagca

Rev 5'-3': ccaaactatgtgataataaatggg

M206 = UTY Intron 2b (1221+3671) (541 bp) T to G at position 31.

GtataatactgtggttggaaagcaCTAAAAKTTAATTTTGGCTTACAGCATTATGCCTATAA ATAAATTTTGCCACCTGAGTCACAGACAAAACAGGCAAAACAATCTTATTTG GCAATTTAAATAATATCAAATGTTCCCTAGTTATTTCAATTTGACTCTTTTAAA AGCTAGCTAGTTAGTAATAAAAGTAGGCTGGATGCAGTGGCTCACTCCTGTA ATCCCAGCACTTTGGGAGGCTGAGGAGAGCAGATCACCTGAGGTCAGGAGTT CCAGACCAGCCTGGCCAACATGATGAAACCCTGTCTCTACTACAAATACAAA

For: 5'-3' = gtataatactgtggttggaaagca Rev 5'-3': ccaaactatgtgataataaatggg

For: 5'-3' = aggaaaaatcagaagtatcctg Rev 5'-3': caaaattcaccaagaatccttg

M208 = UTY1 = Intron 3b (1330+5798) (507 bp) C to T at position 352.

AtaaatacaaaatcacctgatggatATGCAAAAATTTATCAGCTTTACAAAGACATATAATA CCATTCTATGAGCACAAGTTTATTGCAATATTTTGTCCTTTACTGTCAACAAA AGAACACAGCCACATGATATAGGAAAAATCTATATTCTTTACAAATTTTCCAT GAATCTCTAGCTAAAAGATCATATGACATATATGCAACGATTTATCAGCTTTC AGAGCTTTAATTGATATTCATTACTTGTGGGTTCTGTTATTTGACTCACGAAA ATTTATATATACACAAAATCAATACTTAATGATGGTTTCAAAGATATTCACAG ACCTGCTCAGGGCAGCAATAAATTYGACCCACTGGATACACACACTCCCAGCTA ATGTTAGAAGCGGTGGGCCTTTCTCTGACTTCATGTGTCAAGTATTCTAAACA AACAGGCTTTTCCTGCTGTATGCAGTGTCACATTTTTCTGATTTTTGCTCttttgtta gtaatttcgctgtttaa

For: 5'-3' = ataaatacaaaatcacctgatggat Rev 5'-3': ttaaacagcgaaattactaacaaaa

M209 = UTY1 = Intron 3c (1330+6211) (550 bp) A to G at position 471.

CactgtcttccacaatggttgAACTAGTTTACAGTTCCACCAACAGTGTATAAGTTTTCCT
ATTTCTCCATATCCTCCAGCACCTGTTGACATTACTAAAATAACATTCTCAT
CAAGGTCATCAGGGTCTCAGAACTGGCTACATACAACCTCCAAGAAAGTTTC
GTTCTTTCTGTTTTTGCAATGTGTTCTGCCACAAATTCATCAGTTCTCAAAGCT
AACAGAACTTTTACTAGTTGCCCAATGCATCAATTCCATAGTTCTGAGAGCAT
GGGCATGAATGTCTGAAAAACCTGAGGTATGATCACTAATATGCTATTCTCTGA
ACTTCTCAATTGCATTTTCCTCCTTGAATAAATCAGACTAAATTAGTGACACC
ACAAATTGTGATCATTGAGAAATCTCTAAAGGTTTTTCAGAAGCCGAGTAGG
AAGCTATCTATGACTTTTTAAAACTCTGACTGAATTCTRAATATTTAATTG
GACATTACATGAAGACGTTGTGTATTTAACTTCTGAATGCAgggaagataaatacaaaat
cacct

For: 5'-3' = cactgtcttccacaatggttg Rev 5'-3': aggtgattttgtatttatcttccc

M210 = UTY1 = Intron 3d (1330+6221) (550 bp) A to T at position 461.

CactgtettecacaatggttgAACTAGTTTACAGTTCCACCAACAGTGTATAAGTTTTCCT
ATTTCTCCATATCCTCTCCAGCACCTGTTGACATTACTAAAATAACATTCTCAT
CAAGGTCATCAGGGTCTCAGAACTGGCTACATACAACCTCCAAGAAAGTTTC
GTTCTTTCTGTTTTTGCAATGTGTTCTGCCACAAATTCATCAGTTCTCAAAGCT
AACAGAACTTTTACTAGTTGCCCAATGCATCAATTCCATAGTTCTGAGAGCAT
GGGCATGAATGTCTGAAAAACCTGAGGTATGATCACTAATATGCTATTCTCTGA
ACTTCTCAATTGCATTTTCCTCCTTGAATAAATCAGACTAAATTAGTGACACC
ACAAATTGTGATCATTGAGAAATCTCTAAAGGTTTTTCAGAAGCCGAGTAGG
AAGCTATCTATGACTTTTTAAAACTCTGWCTGAATTCTAAATATTTAATTG
GACATTACATGAAGACGTTGTGTATTTAACTTCTGAATGCAgggaagataaatacaaaat
cacct

For: 5'-3' = cactgtcttccacaatggttg Rev 5'-3': aggtgattttgtatttatcttccc

M211 = UTY1 = Intron 4a (1381+16283) C to T at position 381.

For: 5'-3' = caattcactatttgaggaatcca

Rev 5'-3': gaagtctctgatttatttggcag

For: 5'-3' = tataatcaagttaccaattactggc

Rev 5'-3': ttttgtaacattgaatggcaaa

M213 = UTY1 ex05b = Intron 4c (1381-78) T to C at position 290. Mimics M89 (409 bp); non coding

For: 5'-3' = tataatcaagttaccaattactggc

Rev 5'-3': ttttgtaacattgaatggcaaa

M214 = UTY1 ex12 = Intron 11 (1971-60) (460 bp) T to C at position 404; non coding TattacaaaatatggaaacaaggcAACATCAAAACACAAATAGACAAACTTGCCAGCCACC CTTCTCCTGCCAATTATTATAGGAATATACGTGTCATTTAAAATATACTATTT AAAATTTTTACCTGTAGAAAATTTAATTCTTGCAGCAAGCGTAGAGGTATTACT ACAACGTTTGCTTCTAGCTGCATTTAGGTAGCATTTAATGGCATCTTGAGGTT GATTGCAGGATTCATAGAGAGTACCTAGGTCCATCCAGGCTGCGGCATGCCC ATGGTCCAATTGTACAGCACAAATATATGCCTGTAAAGCATCCATAGGCTGA TTTTGCTGCTGATACAACACACTGGAAAGAAAAAGAATGCTGTCAAAAAACTA CTGGTTACTTTCGTTCGTTTATTTTTCYGTTGTTTTCAGACAGTGTCTCACACT GTCTCCCAGGCtggagtgaagtggcatttc

For: 5'-3' = tattacaaaatatggaaacaaggc

Rev 5'-3': gaaatgccacttcactccag

M215 = UTY1 exon 14 (2358) (386 bp) A to G at position 163; silent substitution, SER GtaaaactcagatatacatcccatgAAATATACACAGAAACTATAAATTAGCATTAATATC CTCTAAAATGATACTGTAGTAAAGAAAATATTCTCAAACTGTTGGTAAATTTTA GAGAAAATAAAAAATATTATACATACTTGCTGCATTAAGACAAACTGRCTTTC TAACTGTTCCAGCTGATGCTTCTGTGCTGGATTTAAATTATCTCTATTTGCTCG CAGTTGTTCCAAGTGCTAGAAGAAAAAGAGATTAATAATCAAAGTTTAATC TAAAATTTAAGACAATATAAGGCAACTCCTCACTAAAAAAGACTACACAGAAC CTTTGCAGGATGAAAAGACAGTGATTCCTAATGAACgttaagatagtgattctttttttt

For: 5'-3' = gtaaaactcagatatatacatcccatg

Rev 5'-3': aaaaaaaaagaatcactatcttaacg

 $M216 = UTY1 \text{ intron } 18\ 3678+537\ (557\ bp)\ C \text{ to } T \text{ at position } 54.$

TGCTCCCTAAGGCAATCTGGCTTGATTTAAAGAATACATCGATTTTCTacaagaca cattagttcagactctc

For: 5'-3' = ctcaaccagtttttatgaagctag Rev 5'-3': gagagtctgaactaatgtgtcttgt

 $M217 = UTY1 \text{ intron } 17\ 3678+768 \ (461 \text{ bp}) A \text{ to } C \text{ at position } 219.$

For: 5'-3' = gettatttttagtetetetteeat

Rev:5'-3': acctgtt.gaatgttacatttcttt

 $M218 = UTY1 \text{ intron } 16\ 3679-281+768 (482 \text{ bp}) \ C \text{ to} T \text{ at postion } 380.$

For: 5'-3' = ttgtgagtttttttccatcaatc

Rev 5'-3': tttattgacgatggtattagaagag

 $M219 = UTY1 \text{ intron } 16\ 3676-294\ (482\ bp)\ T \text{ to } C \text{ at postion } 232.$

TtgtgagtttttttccatcaatcTGGCTATTAAAAATCTGCAGTGCATCCTAACCTTTGATAT
TATGTTGCTACATATTACAGTATTGTATCATTTGTCTTGTCAGGAAAGTGTGG
AGGTAATAGCTAAAAAAAAACCCTCTCTTTTAAAAATTACATTTTAAAATTTGAT
TCACTTTAAAACTGTTACCTATCTCTTATACCACAGTGATTTATAAAATTCTTT
TAAATTAGYTGAGTTGTTCGAAAGTATTTCCCAAGCATATTTTTTGAGTTATC
TTCTATTGCTTCTTAAATGAGACAACAGGTAGAAGAGACATTTAAAGTTTAA
AATCAAACTGTTTTATAAACTATTAACAAAACTTTTAGGGAATAAAAACCAC
AACAGGCAAACCTTAAATTTGTATTTATTGCCTCAAAGTTTCAACTGAAACGC
TTATTTTAGTCTCTCTCCATGActcttctaataccatcgtcaataaa

For: 5'-3' = ttgtgagtttttttccatcaatc

Rev 5'-3': tttattgacgatggtattagaagag

 $M220 = UTY1 \text{ intron } 16\ 3676-329\ (482\ bp)\ A \text{ to } G \text{ at postion } 367.$

TtgtgagtttttttccatcaatcTGGCTATTAAAAATCTGCAGTGCATCCTAACCTTTGATAT
TATGTTGCTACATATTACAGTATTGTATCATTTGTCTTGTCAGGAAAGTGTGG
AGGTAATAGCTAAAAAAAACCCTCTCTTTTAAAAATTACATTTTAAATTTGAT
TCACTTTAAAACTGTTACCTATCTCTTATACCACAGTGATTTATAAAATTCTTT
TAAATTAGCTGAGTTGTTCGAAAGTATTTCCCAAGCATATTTTTTTGAGTTATC
TTCTATTGCTTCTTAAATGAGACAACAGGTAGAAGAGACATTTAAAAGTTTAA
AATCAAACTGTTTTATAAACTATTAACAAAACTTTTAGRGAATAAAAACCAC
AACAGGCAAACCTTAAATTTGTATTTATTGCCTCAAAGTTTCAACTGAAACGC
TTATTTTTAGTCTCTCTCCATGActcttctaataccatcgtcaataaa

For: 5'-3' = ttgtgagtttttttccatcaatc Rev 5'-3': tttattgacgatggtattagaagag

M221 = UTY1 intron 18 (3784+165) (324 bp) G to A at position 200.

For: 5'-3' = gggaaatgtgaaaggaaaata Rev 5'-3': ttaaacttttataacactgacagaaac

M223 = A8.05e (208 bp) C to T at position 67.

ttcagcaagagtaagcaagaggCACTGAGCCGCTGGAGTCTGCACATTGATAAATTTACTT ACAGT**Y**GTAAATAAATTGCATCATCTTCAGCTAGTAACACAGAGTCTAATTT TTATAGCGGCATACTTGCCTCCACGACTTTCCT**A**GACACCAGAAAGAAAGGC **G**AGAGCCAGCCTTAGCCTAATCaagaaccatgatccaaaaagg

For: 5'-3' = ttcagcaagagtaagcaagagg

Rev 5'-3' = cctttttggatcatggttctt

M224 = B9.60b (301 bp) T to C at position 193

CttcaggcattattttttttggtTCTCCACTACAGGAGAAATGTAAATGTGATGAGTCAGAAT TTAGGATGGCTGTATGGGTTTCTTTGACTAATACAAGAAATCACTTTGTAATG AATGAAATCAGTGGTTTCTGCATTACTCCGTATGTTCGACATGAACACAAATT GATACACTTAACAAAGATACTTCTTTCYGCCCTTCCAAATATTTCAAAATAAG CTGGTCATAGTACTTGCTTTTCATAAAAAAGATGGTAAGCTTCCAATATTTAGA TTTaaggaaaggtg:aaggaacactat

For: 5'-3' = cttcaggcattatttttttgg

Rev 5'-3' = atagtgttccttcacctttcctt

TTTGTTGACTTACGTCAGGCATTGCTCCACTGTACTCCTAGGCTGCTGGGACC CCGCCCAGCCAGTTCGCCAAGGACCTAGGAACATGACAGAGGCTGACT**R**ATT CTGACCGCTGGTTGGTTGATGGTCACGTCTATGGAGAAAAGGGTAGTCTCTG GGATGGAACAACCTGTAGGTTGTGCTAGTTAAATGCATTAAGATAGAAAATG GAGTGTCTGTGCTGGGTGTTTTTGCAGTTGCGatacgcttgaaggggaagag

For 5'-3'= aaggaaaaagctgaggca

Rev 5'-3'= ctcttcccttcaagcgtat

M226 UTY Ex1c 1104 silent/glu (380 bp) C to T at position 158

For 5'-3'=gagtgccaagctgaggatg

Rev 5'-3'=aaattctgctgtgctctcca

M227 UTY Ex1c 1105 Glu/Gln C to G in at position 157

For 5'-3'=gagtgccaagctgaggatg

Rev 5'-3'=aaattclgctgtgctctcca

M228 UTY Ex1c (380 bp) 1106 Glu/Gly T to C at position 156

For 5'-3'=gagtgccaagctgaggatg

Rev 5'-3'=aaattctgctgtgctctcca

M229= UTY1 Int12, A to C at position 159. (1560+7060 T to G in intron6) Group I

GgtacacacctgtagtcccaacTGCTTGGGAGTCTGAGATGGAAGGATCACTTTGGGCCAGGAATTCCACGCGTTGTACTATGATTATGCCTGTGAATAGCCACTGCACTCAATCCTGGAAAACAGTGAGAGCCAGTCTCTTAAAGTATAATTTCCTTMAATAAAAT

ATATTTCAAAATCTCTCATTCTTATTTATGATCAAAAAATGTTATTCATCAATG
TAGACTTTGAGCTTGGTCAATACTGAGCAAATAAAGCCCTCAAATATCCTTTT
CATTTGACAGGTAACTACATGCCTACTAAGGCCACGTATTATGCATATAACAA
TAAACAAACATAATCCCTCCACGAAAAAGCTCCAGCCAGAGAGAAATATTAA
AGTAAATAATTATGCTCATCTAATCCATTCAGCAATGGCAAGAATTTCACATG
AAAGTACAAGATGTCCAGCACAGATCTAACCACCTACAAATGGATGCCTCCTT
GAGAAAATGTTATTAAGGTAGGACCTGCATGGATAAGTAAAAGttaccatgaaagagtt
ctaaaaaatg

For 5'-3'=ggtacacacctgtagtcccaac

Rev 5'-3'=cattttttagaactctttcatggtaa

M230 (449 bp) UTY Ex9 intron 8 1651-143 **T to A** at position 367 Group VIII

For 5'-3'=aatgtcacatttagtcttaacccat

Rev 5'-3'=acattattagtatgtaaatcttcattgc

M231 UTY Ex13 Intron 13 2283+33 G to A at position 110 in Group VIII

CctattatcctggaaaatgtggGCTCGTTTTAATTATTCATATTAATTTAGTTAATCATC ATTCAATTAATACCTAAAAAAACAACATTTACTGTTTCTACTGCTTTC**R**AATTG GGGGAAAGATCGTCAAAGAATTCATACCTGTAATTTCTGTGGTGTCAAACAC AACGAATAAACTTGCTGTACTGGATGATGTGAAAGACTCTGGCCACCATTCC AGTTATCAGAACCATTCTAAGGAAAAATTTAGTGTAAAAGATTAAGAATATTT GCTTAATTTCATACACTTAGAGTTATGACTAGTGAGAAACcaagtgactaggaat

For 5'-3'= cctattatcctggaaaatgtgg

Rev 5'-3'=attccgattcctagtcacttgg

M232 = UTY1 intron 17 3679-566 (461 bp) **C** to **T** at position 38 Group VIII

TAATCACTTTTATATCCTCAACCAGTTTTTATGAAGCTAGAAAAAAATTCCTT

TATTaaagaaatgtaacattcaacaggt

For 5'-3'=gettatttttagtetetetteeat

Rev 5'-3'= acctgttgaatgttacatttcttt

M233 = UTY1 Exon18n, T to C at position150, (3784+37 A to G at intron18) Group III

AtcacttgcatcagtgctaaagaTGCTTGCTCATGCACAAGAGGTATAAAATTGAGTGAGA AAGAAAGATAACACACATTAAAAATAAAAGACTCAGAATGTTGGGGGAAAAAAT CAGTGAGTTTCTGTCAGTGTTATAAAAAGTTTAAAGAYAGTAAAAATATATTTC AATCTTGGTTTTAAGCTTACCTAATTTAAGAGCTCCAGCAAGGCCACGTATTA CTGTAACAGGGTTTTTTGGATttgtacaaaaattgatgtaatggagGAAAGAAAGCATCACGTT TATTTTCCAACTGTAAAAAGCAAAATATTTTGTTAGGTCTCAGATAAAATGACAA AATATACCTCAGATTTGTGCCTTTAATAAAAATGATTAAAATACAATACTTCAAA TTTGTGAGTTTTTTCCATCAATCTGGCTATTAAAAAATCTGCAGTGCATCCtaacct ttgatattatgttgctacat

For 5'-3'=atcacttgcatcagtgctaaaga

Rev 5'-3'=atgtagcaacataatatcaaaggtta

M234= UTY1 Exon20n, C to T at position 253, (4049 G to A in cDNA, codon 1015, Arg/Gln)

Group III

For 5'-3'=tctccattagcaatgtgtttt

Rev 5'-3'=caataccaggetteeteett

M235 = (317 bp) DFFRY Exon4, T to G at position 155. (1859 in cDNA, codon 65, Asp to Glu

For 5'-3'= tagatatttttccttaatctgtggt

Rev 5'-3'= actacaatctactcttttcttttctc

M237= DFFRY Exon30, (366 bp) G to C at position 39. (5903-132 in intron29)

Group III, 325 bp w/out homopolymer region in STS.

For 5'-3'=ttgcatttactgttctagagagttct

newRev 5'-3'=gtacttactttaatgagattagcac Homopolymer clipped off

M238= DFFRY Exon43, C to G at position 28 (8729-54 in intron42) Group I

For 5'-3'=gtactaaatggcacataattaggaa

Rev 5'-3'= aatttagaatgattttcagtatcagc

M239 = DFFRY Exon43, G to A at position 148 (8795 in cDNA, codon 2377, silent/Ser Group I

GtactaaatggcacataattaggaaCTSAAATGTTAGCTACTATTGGATATTACAAAGTTTTACATCTGCTTCTGTTTTAGAATTCATAATGCACTTAAAGGAATTCCAGATGACAGAGTGGGCTGTTCGATACAATACAGCGCTCRAAGAATCACTATCAAAAACGAGCATATCAGTGCATAAAATGTATGGTAGCTCTATTTAGCAGTTGTCCTGTTGCTTACCAGATCTTACAGGTGAGGGTTTTTCTCTTATAAATTTGTAGAAACCTCTGTCACAAGTAAGGAAATGATCGTGAAATTTTTTGTATTAGCATTTTAAgctgatacttgaaaatcattctaaatt.

For 5'-3'=gtactaaatggcacataattaggaa

Rev 5'-3'= aatttagaatgattttcagtatcagc

M240 = DBY int2n, C to T at position 47, (116+613 in intron1.

CtgtggaattcttgaagacgagTGACTATAATATAGCACAACGTAAYAAGTATCCTGTATC
TTGTTTCTGGTGGGGTCCCGTAGCCACGGAGCAACCGTTGCCCGGGTGCTGAG
CGTGCCGAAACTGGGCTTCCGGTATGGAAAGTTTTGTGACGCAGAAGGACCG
GAAAGGGATGGTGGGGAGGGTAGGGAAGGATGGCTGCCGCGTGCTTCTCTTG
ACCCTGTAGAAATAATGGAAATTGGACGCCCGCGGAAAGACACCTGGAAGGT
TAGAGATCCAGCATTGCGCTACACCCCTTTGTTAATTCAGTCACTGGACAGCC
GCCTAGCCGAGAGCTGTGCGGTTTTTATATGGTATTGTATCTTTACTTTAGGCG
ATACATGCAGAAGTCGTCCGGTAgaaaactaacctcgaatgttgatt

For 5'-3'=ctgtggaattcttgaagacgag

Rev 5'-3'=aatcaacattcgaggttagttttc

M241 DBY Intron 4 (intron 1) G to A at position 57 cDNA# 117-989

5'-3' For aactcttgataaaccgtgctg

5'-3' Rev tocaatctcaattcatgcctc

M242 DBY Intron 4 (intron 1) C to T at position 337 cDNA# 117-866 Group X

AactettgataaaccgtgctgTCTAGTTCACTAGAATTAAGTAGTAAATTCAGATGGCAA GATTTTTAAGTACAGTAGTATCTTAATTGATGATTCATGTAATGTGATAGTAT CTTGAACTTATATGTAAGCTTTCTACGGCATAGAAAGTTTGTGCAAAAAAGG TGACCAAGGTGCTYTTGGCATTGGTCTTAACGTGTTTTTTGAAAAAAAATCTAT TTTAACGTACATGGTTTTTTCCCCCACCCCGCCACCGCTTCAGAGTTGTTCTA GGTAAGGTATTATGCTGAAAGCCCTTAAAGCGAAATAACCTTTTTTCTAGTTT TAAAATCCATCAGTATAAGgaggcatgaattgagattgga

5'-3' For aactcttgataaaccgtgctg

5'-3' Rev tccaatctcaattcatgcctc

M243= DBY int6, (401 bp) **T to C** at position 142, (117-356 in intron1) Group III

For 5'-3'=ttttgagcttttgatgtttagga

Rev 5'-3'=caaacttagctgccatgtcg

M244= DBY int6, (401 bp) A to C at position 174, (117-323 in intron1) Group I

CATGGTTGGTAGACCTGGACATCTTTAAAATTTGGCAGGTAATACCAGGCcgaca tggcagctaagtttg

For 5'-3'=ttttgagcttttgatgtttagga

Rev 5'-3'=caaacttagctgccatgtcg

M245= DBY int8, del AAACA at position 264, (174+779 in intron2)

Group I

For 5'-3'=gacgaagaacctaacattcagtg

Rev 5'-3'=tagacagtacttaactacctgattttg

M246= DBY int3, T to G at position 284, (174+799 in intron2)

Group I

For 5'-3'=gacgaagaacctaacattcagtg

Rev 5'-3'=tagacagtacttaactacctgattttg

M247= DBY int9n, T to C at position 224, (175-693 in intron2)

Group II

For 5'-3'= atggtagagacatttttggatattt

Rev 5'-3'=gcctgtaatcccagcacttt

M248= DBY int9n, T to C at position 494, (175-444 in intron2)

Group VI

 $GCCT {\color{red} Y} GCCT {\color{gray} C} CCC a a a gtgctgggatta c a ggc$

For 5'-3'= atggtagagacatttttggatattt

Rev 5'-3'=gcctgtaatcccagcacttt

M249= DBY int10, A to G at position 313, (175-167 in intron2)

Group II

For 5'-3'=tttcaccttgttagccaggat

Rev 5'-3'=caagatactgcttttagaaaatgtct

M250= DBY intl1n, A to G at position 299, (223+687 in intron3) Group III

For 5'-3'=taacagttgttaagattaccactttt

Rev 5'-3'=aacctitgatgcaattccag

M251= DBY int12n,(site a) (nominal, 418 bp) G to A at position 279, (223+1051 in intron3. Site within STS with a 7 T homopolymer length polymorphism allele. aaatattgcatctggctggaATTGCATCAAAGGTTTATTAACTGCCTTAAGGAGAGAGTTGGCAATATTTTAGTATTTGAGGGGATGGAAGAGACCTTAAACATCTAACTTCCTAAATCTGGGAAGTACAATCGATTTAGTACAATAGATCTAGATTTAGGAAGTACA

ATTATTCATTTGTCTAATATTGGAGATTTAAAAGCAGGGGAAAATAACTTTAT TAACTTGTAACTTTAAACATTCATTGAAATGTTTGAAATTTAGGTAAGTGTGTG GTTGTGRAgtgagtttactcttgtcattTTTTTTTATCAGTTTGTAGACATGGAAAGTAG GCAACAATGAGGGTTTTTTTGTTTTAACACACAAGTATACCTTATTCTTAACGAG CATATTaagattacatagttacttttggactt

For 5'-3'=aaatattgcatctggctgga

Rev 5'-3'=aagtccaaaagtaactatgtaatctt

New Rev 5'-3'=aatgacaagagtaaactcac to exclude poly T region

M252=DBY int12n, (419 bp)ins T at position 354, (223+1127 in intron3. (site b) Homopolymer 7T's to 8T's

Group VI.

For 5'-3'=aaatattgcatctggctgga

Rev 5'-3'=aagtccaaaagtaactatgtaatctt

M253 = DBY int 13, (400 bp nominal) C to T at position 283

Group VI

For 5'-3'=gcaacaatgagggttttttg

Rev 5'-3'=cagctccacctctatgcagttt

M254 = DBY int13, (400 bp nominal, 418 bp derived)18bp INSERTION + 2bp substitution, A to G and G to C at positions 339, 340

Group VIII

TGCTACATACTATGTTTACTATGATRSTTGCTACATACTACTATGTATG AGTAGTTTTTCGTCATaaactgcatagaggtggagctg

For 5'-3'=gcaacaatgagggttttttg

Rev 5'-3'=cagetceacetetatgeagttt

M255= DBY int14, (within derived 471 bp) C to T at position 107, (224-813, in intron3) Group V

ttttttttgagacggagtc:ttgCTGTGTTGTCCAGGCTGGAGTACAGTGGCGCGATCTCAGC
TCACTGCAAGCTCCACCTCTTGGGTTCATGCCATTCTCCTGCCTYAGGCTCCT
GAGTAGCTGGGACTACATAGGTGCCCGCCACCATGCCCAGCTAATTTTTTTGT
ATTTTTAGTAGAGACGGGGTTTCACCGTGTTAGCCAGGATGGTCTTGATCTCC
TGACCTTGTGATCTGCCTGCCTTAGCCCTCCCAAAGTGCTGGGATTACAGGT
GTGAGCCATCCCTGTTTTAATCCATCTGACATATTTCTTCTGATTATGTAGCTC
TCTTAGTTCAAGCTTTTCTGTAGGTAACCCACAGTCCCTGAGGTAATCTTTTA
CTTAGCTGGGCCTTCCCAAAATGTGTATTATATATATAGCATATGTTAAATGTTT
AGGTTTAACACCTtttgtattattcaggatttgtcaag

For 5'-3'=tttttttgagacggagtcttg

Rev 5'-3'=cttgacaaatcctgaataatacaaa

M256 = DBY int14, (derived 471 bp) ins \mathbb{C} at position 249, (224-672 in intron3) Group V

ttttttttgagacggagtcttgCTGTGTTGTCCAGGCTGGAGTACAGTGGCGCGATCTCAGC
TCACTGCAAGCTCCACCTCTTGGGTTCATGCCATTCTCCTGCCTCAGGCTCCT
GAGTAGCTGGGACTACATAGGTGCCCGCCACCATGCCCAGCTAATTTTTTTGT
ATTTTTAGTAGAGACGGGGTTTCACCGTGTTAGCCAGGATGGTCTTGATCTCC
TGACCTTGTGATCTGCCTGCCTTAGCCCTCCCAAAGTGCTGGGATTACAGGT
GTGAGCCATCCCTGTTTTAATCCATCTGACATATTTCTTCTGATTATGTAGCTC
TCTTAGTTCAAGCTTTTCTGTAGGTAACCCACAGTCCCTGAGGTAATCTTTTA
CTTAGCTGGGCCTTCCCAAAATGTGTATTATATATATAGCATATGTTAAATGTTT
AGGTTTAACACCCTtttgtattattcaggatttgtcaag

For 5'-3'=tttttttgagacggagtcttg

Rev 5'-3'=cttgacaaatcctgaataatacaaa

M257= DBY int14, (nominal 470 bp) T to C at position 373, (224-547 in intron3) Group I

ttttttttgagacggagtcttgCTGTGTTGTCCAGGCTGGAGTACAGTGGCGCGATCTCAGC
TCACTGCAAGCTCCACCTCTTGGGTTCATGCCATTCTCCTGCCTCAGGCTCCT
GAGTAGCTGGGACTACATAGGTGCCCGCCACCATGCCCAGCTAATTTTTTTGT
ATTTTTAGTAGAGACGGGGTTTCACCGTGTTAGCCAGGATGGTCTTGATCTCC
TGACCTTGTGATCTGCCTGCCTTAGCCTCCCAAAGTGCTGGGATTACAGGTGT
GAGCCATCCCTGTTTTAATCCATCTGACATATTTCTTCTGATTATGTAGCTCTC
TTAGTTCAAGCTTTTCTGTAGGTAACCCACAGTCCCTGAGGTAAYCTTTTACT
TAGCTGGGCCTTCCCAAAATGTGTATTATATATATAGCATATGTTAAATGTTTAG
GTTTAACACCTtttgtattattcaggatttgtcaag

For 5'-3'=tttttttgagacggagtcttg

Rev 5'-3'=cttgacaaatcctgaataatacaaa

M258=DBY int15, (475 bp) T to C, at position 123, (224-388, in intron3)

Group VI

Tatatag catatg ttaaatg tttaag tTTAACACCTTTTGTATTATTCAGGATTTGTCAAGGATGGGACATAACTAAGAAACTAACAATGGGCTTGCACTAGCTACAAGTTCAGCTT CTTAAATAATTTAATTGGAGTAGGTTTATATTTTGGATATGTAACATTTACAC CTATGTTACATGCAAGCTACCTTGTTCTCAGGTCATGAGATTACTTTGCTTCAT ATAATAATCTCTGGTGGAAGAAAACATTAAAGCTTTTAACAATTCTGCTTATG GGACTTGTAGACCATTGGTCCCATAAAGATAACATAAAGGAAGACTACATGT GAAGGACTTCATATTTTgaaagatgcaaattattcaaaagtc

For 5'-3'=tatatagcatatgttaaatgtttaggt

Rev 5'-3'=gacttttgaataatttgcatctttc

M259= DBY int16, (396 bp)T to G at position 151, (352+271, in intron4 Group IX

Caga at gtt ggtttact catt gtt TTGTTAGCAGTAAGAGGTCTTTATTAATTTAATTAAATTAGATGAATATGGTATTTGACACAGTGAAATCTGTTTCAACTTAAATGATACTTA AAGCCTGTCTGTGACAGCTTTAAACACTTCATTT**K**TGATGTGTGTTATAAGTT GATCTTAAAAACCTAATGGCTGTATTTAATCCTTTCTGTTTTTCACAAATAGG AGTAAAACTCTAAAAATATTCTCTTGTCACATGTCTACTTTCATATAAAGGAG AAATTCAAGTGTTATTCCTGCTTTCCTACTAGTAAATATATTTAGATGATACT ATTTTAAATGAAGATGTAAAGTACGTAACTAGTTATAAGTATCTaaaaaacctaattctt agcatgtga

For 5'-3'=cagaatgttggtttactcattgtt

Rev 5'-3'=tcacatgctaagaattaggttttt

M260= DBY int19, (343 bp) G to A at position 253, (608-124 in intron6)

Group VI

CcacacccagctcatttttGTACTTTTAGTAGAGACAGGGTTTCGCCATGTTGGCCAGGC TGGTCTCAAATTCCTGATCTCAAGTGATCTTCATGCCTTAGCCTCCCAGAGTG CTGGGACTACAGGCATCAGCCACCATACCTGGCCTCCAAAAACTTTTTTCAAT GTAGATTAAACCCAGGCATTTTCTTAAAAAATGCCATGAATCTTTTACTGAAA TCATAGCATCTGTAAACTAAATCAGACAGTTTA ${f R}$ TTGGTTACTTCCATTAATA TGTTAGTATAAAACAGAAATTGCGACAGATACAGCATTTTATATCtgctatgtttacttc tgtatttactt

For 5'-3'=ccacaccageteattttt

Rev 5'-3'=aagtaaatacagaagtaaacatagcag

M261= DBY int22, (284 bp) A to G at position 213, (1090-32 in intron10)

Group X

AtttgaggctctgagcttcaTTTTAACAATCAACATGGGTAATTCGGTTGTTACCTTGAGC ATTTCATCTCATGATTTTGTGTGTGTTTTGTGTGTGTATGCATTTGTTGAGTATA TGTCAAATTGTGACACTGCAATAGTTACTACTTGAGTTACTATATTAGTGCAA

TTAATTACACAACTATATATAGTAATTAGTTTCTCAGATCTAAT**R**ATCCAGTA TCAACTGAGGGTTTTCGTAATAGGTACTTAGTGTTGGATGAAgctgataggatgctggat atg

For 5'-3'=atttgaggctctgagcttca

Rev 5'-3'=catatccagcatcctatcagc

M262= DBY STS01, (502 bp) del A at position 226, (1-2908 out side of 5' region) Group III

agctgtttggacttgagtagttgTAGAATAACTGAAAAATAGGAAACTGCTATATATATATGT
ATGTATAATATATATAACCTTTTTTCAGGTACTCCTATTGCAATACCTGCATTT
CAGCACTATTCAAAAAGTAAAATAAGTCCCAGAGCCAGGTTAGTCATTATGTC
CTATTTATTGCTAATTTTCATATACAAATGAGAGCTGTCAGAATTCACAGCTT
CTGAATATCAGAAGCTCATGTTTTCCCTGGTCTATACAAAAAGGAAATAAGT
GAGGCCAAAAATGTACTTTAACAGTGCTCCATAATACGAATCTCATAAATGA
GCTGGAATAGACCCTGAGGTCTTCAAGCCTAGTTTCTCAAGATCGTATTTTGT
AAACTTGTGCTAGCAGTTTTGAATATCACAATGATTGGCATGGGCTGCTGACA
TTTTAGCAGGCAGGGCTCAGGGTGTTAGATGTCCTGTAATTCAGGgacattcacagta
gaaaatactttgg

For 5'-3'= agctgtttggacttgagtagttg

Rev 5'-3'= ccaaagtattttctactgtgaatgtc

M263=DBY STS06, (515 bp) G to C at position 332, (1-341 out side of 5' region) Group III

For 5'-3'= ccactcagctttcctcaggt

Rev 5'-3'= cgttcgtttagtcataagatcg

M264=DBY Exon17, (552 bp) C to T at position 115, (1988 at cDNA, codon639, silent/Gly)

Group III.

tccaactctagatttcttttactggTTTTATGTTAAAGTACTTGAGAAAAAAAAAAGGTATTAAC GAATGACTTAATTTCTCTCTAAACATTTTTCTTGATAGGTGGCTATGGAGGYT TCTACAATAGTGATGGATATGGAGGAAATTATAACTCCCAGGGGGTTGACTG GTGGGGCAACTGAATCTGCTTTGCAGCAAAGTCACCCTTACAAAGAAGCTAA TATGGAAACCACATGTAACTTAGCCAGACTATATTGTGTAGCTTCAAGAACTT GCAGTACATTACCAGCTGTGATTCTCCTGATAATTCAAGGGAGCTCAAAGTC ACAAGAAGAAAAATGAAAGGAAAAAACAGCAGCCCTATTCAGAAATTGGTT For 5'-3'=tccaactctagatttcttttactgg

Rev 5'-3'=ggtatcgtgagcattactgagc

CTAAATTAAGTTCTGACTTGCTMACTCTATCCTGGATAGGCACTTGGGAACTT ACACTCTTTAAGCCATTCCAGTCATGATGAGGTGGAATGTATCAGTATACCA ATTAATATTTTTGAAAGAGCTCTTTTAGGTTAATTTAAGTacagcaatttctcatgtaatgttt

a

For 5'-3'=ttagacaacttactactttgatgtcct

Rev 5'-3'=taaacattacatgagaaattgctgt

M266= DBY STS08, (444 bp) **T to C** at position 208, (2312+623 outside 3' region) Group II

For 5'-3'=tgaggtggaatgtatcagtatacc

Rev 5'-3'=tgatttcaaggatttgttagtctt

M267 EIF1A Y STS12 (site a) (287 bp) T to G at position 148. STS also contains two Group I associated mutations

newFor 5'-3=ttatcctgagccgttgtccctg

Rev 5'-3'=tgtagagacacggttgtaccct

 $M268 = EIF1A_Y STS5a$, (427 bp) A to G at position 292,

GROUP VII

For: 5'-3'= ctaaagatcagagtatctccctttg Rev: 5'-3'= actatacttcttttgtgtgccttc

 $M269 = EIF1A_Y STS5b$, (427 bp) **T** to **C** at position 358, Group IX

CtaaagatcagagtatctccctttgCAAAATGTCCATTAAATCTTTGCTGATGTTATTATCCC TGTACCTGACTCTATCCTTAAATAGTAAGGCTTCCTTTATTCTTGTAGGGTAG AACTTTTAAACTGAGTGATGCCTAAAAATGTTCTCAATAAAGAGAGTATCTCC AAAACACGTCGGATTTGTTTAAAGAGGAAGTGTGGATTTTTTGATCTTAGAA AGGAAACGAGATAAAATATTAAACGACTTTAATTTTTGTATGATCATGCCTA GCCTCATTCCTCTAAAATATAATTTAAAGTGGATTCTGTTACATGGTATCACA ATAGAAGGGGAATGATCAGGGTTTGGTTAATYCTGGTAAAATTGAAAACAATT

For: 5'-3'= ctaaagatcagagtatctccctttg Rev: 5'-3'= actatacttcttttgtgtgccttc

M270 = EIF1A_Y STS5, (428 bp) ins T at position 387.. Has ancestral T at M281. HOMOPOLYMER

CtaaagatcagagtatctccctttgCAAAATGTCCATTAAATCTTTGCTGATGTTATTATCCC TGTACCTGACTCTATCCTTAAATAGTAAGGCTTCCTTTATTCTTGTAGGGTAG AACTTTTAAACTGAGTGATGCCTAAAAATGTTCTCAATAAAGAGAGTATCTCC AAAACACGTCGGATTTGTTTAAAGAGGAAGTGTGGATTTTTTGATCTTAGAA AGGAAACGAGATAAAATATTAAACGACTTTAATTTTTGTATGATCATGCCTA GCCTCATTCCTCTAAAATATAATTTAAAGTGGATTCTGTTACATGGTATCACA ATAGAAGGGGAATGATCAGGGTTTGGTTAATTCTGGTAAAATTGAAAACAATT

 $TTTTTTT{\color{blue}TATCATATGTGCCTCA} agaaggcacacaaaaggaagtatagt$

For: 5'-3'= ctaaagatcagagtatctccctttg Rev: 5'-3'= actatacttcttttgtgtgccttc

M271 = UTY1 intron 17 3679-566 (461 bp) A to C at position 296
Group VIII. Discovered while typing M232. This STS also contains M217 site.
gcttatttttagtctctcttccatGACTCTTCTAATACCATCGTCAATAAATTTCAACTAGGTA
AAAAATTAATATGAACATCTGTCCAAAGAAAGGCCAGTATCTCCAAAATCC
TCTCGTACAGATCTGTTTCGAGATCATTCTAATTACTGTATCTTCATATTTTAG
GTTAAGATTCTTTAACTTGTGAAGGAGAATGAAAAAGTTGGGTGACACAAAC
TCTTCAGAAGGAAAAAAAAATTATTTTGATGAAAGCCACAGCAGCT

TTATCAAATGCTTACGTTGCT**M**AATAGTAAAAAAAAGCCACTTAAATTCCAAT GGAAATTTTATACCCACATGTATTTATGTAAAAACTTTTAAATAACATGTATTC ATAATCACTTTTATATCCTCAACCAGTTTTTATGAAGCTAGAAAAAATTCCT TTATTaaagaaatgtaacattcaacaggt

Rev:5'-3': acctgttgaatgttacatttcttt

M272= EIF1A_Y STS4, (496 bp) A to G at position 212, GROUP VIII

Rev 5'-3'=cagcaaagatttaatggacattt

M273= EIF1A STS8, (502 bp) C to G at position 189 GROUP II

CacatcaggaaaagggcatcCTTTGGCCTATACTTGTGAAGAGCTAGAGTAAGGTGCTC
CCCACCTTTGAGATTGCTAAAGTTGTCATTCTTTTTGGAAATTTATGAGCTAAT
CATCATTTAGTCATTTGAAAAAGCTGCCAAACTTTTGTAAAACCCAGTAAGGA
AAGCAGGTATGATCTTTGTCCTGASGCAGCTAAGTTCAGGCACGATTAATTGC
TCGAAATATAGAATGTGTTTTCCTTTGTAGAAATTTAGTTTTGGCATGCCCTA
AAATGCATCAGAATCTGGATAAATCACAGAGTTCTGGAAGCCCAATTGTCTT
CTATAGTGGCACAGAACAATGTGAGACTGCCCCAGAGGTAGTGGGTGAATTC
AAGAAGTTAGATGTCTGGCTTTATGGTGGCCAGGTATATGTTTTATTCTATTT
GCAGTGTTAACATTTTTATTCAAATTCTTCAATCGATCCCTTAATATTACTGTA
atttgtagectttctccctcc

For 5'-3'=cacatcaggaaaagggcatc

Rev 5'-3'=ggagggagaaaggctacaaat

M274= EIF1A_Y STS2a, (457 bp) C to T at position 47, GROUPVIII w/M11

For 5'-3'=gccatgcccaagaataaag

Rev 5'-3'=ctaaacatgcttcaaggcttc

M275= EIF1A_Y STS2b, (457 bp) C to G at position 325 GROUP X

For 5'-3'=gccatgcccaagaataaag

Rev 5'-3'=ctaaacatgcttcaaggcttc

M276 EIF1A_Y STS12 (site b) (287 bp) T to A at position 58.

Group I associated mutation. Has another Group I site (M277) and a Group VI site (M267).

newFor 5'-3'=ttatectgagecgttgtccctg

Rev 5'-3=tgtagagacacggttgtaccct

M277 EIF1A_Y STS12 (site c) (287 bp) G to T at position.

Group I associated mutation. G to T at position 151. Has another Group I site (M277) and a Group VI site (M267).

newFor 5'-3=ttatectgagecgttgteeetg

Rev 5'-3'=tgtagagacacggttgtaccct

M278= DBY int12n, site c ((nominal, 418 bp)) T to G at position 374, Site within STS with 7 T homopolymer.

Group I.

a a a tatt g catct g g c t g g a ATTGCATCAAAGGTTTATTAACTGCCTTAAGGAGAGTTGGCAATATTTTAGTATTTGAGGGGATGGAAGAGACCTTAAACATCTAACTTCCTA

AATCTGGGAAGTACAATCGATTTAGTACAATAGATCTAGATTTAGGAAGTAC
AATTATTCATTTGTCTAATATTGGAGATTTAAAAGCAGGGGAAAATAACTTTA
TTAACTTGTAACTTTAAACATTCATTGAAATGTTTGAATTTAGGTAAGTGTGT
GGTTGTGGAgtgagtttactcttgtcattTTTTTTTTATCAGTTTGTAGACATGGAAAGTA
GGCAACAATGAGGGTTTTTTTTGTTTTAACACAAGTATACCTKATTCTTAACG
AGCATATTaagatlacatagttacttttggactt

For 5'-3'=aaatattgcatctggctgga

Rev 5'-3'=aagtccaaaagtaactatgtaatctt

New Rev 5'-3'=aatgacaagagtaaactcac to exclude poly T region

M279= DBY int12n, site d ((nominal, 418 bp)) C to T at position 93, Site within STS with 7 T homopolymer.

Group I

For 5'-3'=aaatattgcatctggctgga

Rev 5'-3'=aagtccaaaagtaactatgtaatctt

New Rev 5'-3'=aatgacaagagtaaactcac to exclude poly T region

M280 revised B9.36 c (386 bp) STS G to A at position 280

Group VI

ccagtcagcagtacaaaagttgACAGCTTCAGCAAAATTGTAGCCTTGGTTAAAAACCACTG TGGTAAGCACGAGGAAAAAGTGATGACAAACTCCCCTGCACACTGGTTTGTGC GGACAACCTAAAAAGGAGAAAAAAGCAGAAAGAGGTGTGGGTCAGAACTAA TGGGCCAGATGTGAACTCAAAGATGTCTCTAGATGCTGTAACAGATGTAGGA AGAGTGGAAAGGCTCTATCTTCAAGTACGTGTCCTAAAAGAAAATGAGATTG TGAATTTAAAARTGGTATTCATAGAAAAGTACTCAAAATATGTGTAATTCAA AAAACAAATATAGAGGGGTCCACGAACAAGTGAAAAGACTCTttgcttctataatcaaa gaaatgc

newFor 5'-3' = ccagtcagcagtacaaaagttg

newRev 5'-3' = gcatttctttgattatagaagcaa

M281 = G3.27f (393 bp) G to A at position 247.

Discovered while typing M123

tggtaaactctacttagttgcctttTGGAAATGAATAAATCAAGGTAGAAAAGCAATTGAGAT ACTAATTCATGCTCTCAGGGGAAAATCTGAATAAAGCTATCTTTTCTAACACA GAGCAAGTGACTCTCAAAGTCACAGTATCTGAACTAGCATATCAGCATCGCC TGAATACCTAGAAATGCAAATTCCTGGGCAACACCAGAATCTAACAAAGCAA AAAACTATGGGGGGAACAGGGAAGTCRGTTTAATAATACTGAGTTTGTGCA ACCTCAACTTTGCTTTATAGGAAAGCAAAATCTCAATATGATAAAGTTTTCTT CAACAAAACTCTGAGATAACTATGTTGAGGGAAAGAAGTTGATCACATgcaaga aaatctaattcgctg

For = tggtaaactctacttagttgccttt

Rev 5'-3' = cagcg:aattagattttcttgc

M282 = G3.27g (393 bp) A to G at position 316.

Group VI

tggtaaactctacttagttgcctttTGGAAATGAATAAATCAAGGTAGAAAAGCAATTGAGAT ACTAATTCATGCTCTCAGGGGAAAATCTGAATAAAGCTATCTTTTCTAACACA GAGCAAGTGACTCTCAAAGTCACAGTATCTGAACTAGCATATCAGCATCGCC TGAATACCTAGAAATGCAAATTCCTGGGCAACACCAGAATCTAACAAAGCAA AAAACTATGGGGGGAACAGGGAAGTCGGTTTAATAATACTGAGTTTGTGCAA CCTCAACTTTGCTTTATAGGAAAGCAAAATCTCAATATGATAA ${f R}$ GTTTTCTTC AACAAAACTCTGAGATAACTATGTTGAGGAAAGCAAAAGCAAAGCAAAACTCTGAGATAACTATGTTGAGGGAAAGAAGTTGATCACATgcaagaa aatctaattcgctg

For = tggtaaactctacttagttgccttt

Rev 5'-3' = cagcgaattagattttcttgc

M283 = DBY STS 09b (429 bp) A to G at position?

STS also contains M200.

ggettacacttgcagactttgCAAATCTTAAGACTAACAAATCCTTGAAATCACACAGCTT GCAAATACGTACTAAACTGCACAAGGTGTGTGTTCTATATGTGCAGTTTTAGC GTATTTTAGTTGCATAGGTTTCCATGGTATTTATAGTCTCTTGTGCTAAATTTG GCCAAAGATGATTGTCCACCACTAAAAATGCCTCTCCCACTTGGAATTCTGTA CTGATTTTGTGGCCAGATGCAATGATCTTTAAAAACAAATCTTTTCAATGGCA TAAGAAGTTGACRAAAATTTCTTAAAGTGCAATAGATTTTCAAGTGTATTGTG CCTTGTTCTAAAACTTTTAAGTAGGTGCACTTGACAGTATTGAGGTCATTTGT TAAGGTGCTATTTCAATTAGTGTAggtttagactcttgtacatttctcc

For = ggcttacacttgcagactttg

Rev: 5'-3'= ggagaaatgtacaagagtctaaacc

M284 = EIF1AY STS34a, (399 bp nominal) del ACAA at position 105, STS has another marker, M306,

GroupIX.

Ggaagtgctgaaagtttcgct F 5'-3' = ggcagttttcatttaagcaga

R 5'-3' = agcgaaactttcagcacttc

M285 EIF1A_Y STS12 (site d) (287 bp) G to C at position 70

(GroupVI)

newFor 5'-3'=ttatcctgagccgttgtccctg

Rev 5'-3'=tgtagagacacggttgtaccct

M286 EIF1A_Y STS12 (site e) (287 bp) **G** to **A** at position 129. (GroupVI)

newFor 5'-3'=ttatcctgagccgttgtccctg

Rev 5'-3'=tgtagagacacggttgtaccct

M287 EIF1A_Y STS12 (site f) (287 bp) A to T at position 100. This is one of 3 M201 related mutations.

(Group VI)

 $ttatcctgagccgttgtccctgTGTTTCCATTTCTCTTTTCCTCATTTCTCATCATCTACATTT\\ CTCCTGTACTTGTTCATTAAATAATGATTCCTTGGWTATACCAAGTCTGGAT\\ AGCGGATTCGATGGAAGCATTTTTGTAAATATACGTTCAGTATTTTGTGTGGA\\ AGAACACAATCTAGCTGATGCCTGCAATCCCAGCCCTTTGGAAAGCGAGGTG\\ GGTGGATTGCTTGAAGCTACGAGTTTGACACTAGCCTGGGCAACagggtacaaccgt\\ gtctctaca$

newFor 5'-3'=ttatcctgagccgttgtccctg

Rev 5'-3'=tgtagagacacggttgtaccct

M289 = B9.36new d (386 bp) G to A at position 227 Group VI.

For 5'-3' = ccagtcagcagtacaaaagttg

Rev 5'-3' = gcatttctttgattatagaagcaa

M290 = B9.36new e (386 bp) G to A at position 343. Group III

ccagtcagcagtacaaaagttgACAGCTTCAGCAAAATTGTAGCCTTGGTTAAAACCACTG TGGTAAGCACGAGGAAAAGTGATGACAAACTCCCCTGCACACTGGTTTGTGC GGACAACCTAAAAAGGAGAAAAAAGCAGAAAGAGGTGTGGGTCAGAACTAA TGGGCCAGATGTGAACTCAAAGATGTCTCTAGATGCTGTAACAGATGTAGGA AGAGTGGAAAGGCTCTATCTTCAAGTACGTGTCCTAAAAGAAAATGAGATTG TGAATTTAAAAGTGGTATTCATAGAAAAGTACTCAAAATATGTGTAATTCAA AAAACAAATATAGAGGGGTCCAYGAACAAGTGAAAAGACTCTttgcttctataatcaaa gaaatgc

newFor 5'-3' = ccagtcagcagtacaaaagttg

newRev 5'-3' = gcatttctttgattatagaagcaa

M291 = EIF1AY STS16, (480 bp) A to G, at position 358,

(Group III)

cggagtctggctttgttggcCAGGTTGGAGTGCAGTGGCATGATCTCGGCTCAGGGCAAT GTCCGTCTCCTGGACTCAAGCAGTTCTCCTGCCTCAGCCTCCCCAGTAGCTGG GATTAGAGGTGTGACACCATGCCCGGCTAATTTTTGTATTTTTAGTAGAGA TGGGGTTTCACCATGTTGGCCAGGCTGGTCTCGAACTCCTGACCTCAGGTAAT GCACCCGCCTCGGCCTCCCAAAGTGGTGGGATTATAGGCGTGAGTAACCATG CCTGGCCTTTCACTCTTATTTTCTAAGAACTTTAGAATAATCACCGAGATATT CTAAAGTAAACAGGAATTTTTAATGGTTAAGCTRTTATTTGTCTTTGTCATTTC TGAGTTTAGGGATAGTGAAGATAGAGTTAGGCCTCATGTGTGAGAGACTGAT GTAGCATTATAGTGTATATTTTGAAATGTGccaccgtgatgttcaaaagt

For = cggagtctggctttgttggc

Rev 5'-3' = actttl.gaacatcacggtgg

M292 = EIF1AY STS19, (556 bp) **A to G**, at position 373.

Group III

Original F 5'-3' = tttaacaaatgtggaccaaga

Rev 5'-3' = acttttgaacatcacggtgg

M293 = EIF1AY STS20a, (507bp) **T** to **G**, at position 299.

Group III. STS also contains M294

CatggtccaagcaatttatttttgTGAGTTCCCAAAATAATTTATACAGCAATGATTCATGTG ACAATGTGAATAAATAGAAAAAGTCTTTGATAACTTTTAGATTTACTTTTAAA GAATAATTTGTTTGATTAACTTCTGTTGTATTCCTACCAGAAATGTTTACTCTG F 5'-3' = catggtccaagcaatttatttttg

R 5'-3' = gctggctaatacttccacagag

M294 = EIF1AY STS20b, (507bp) **C** to **T**, at position 305

F 5'-3' = catggtccaagcaatttatttttg

R 5'-3' = gctggctaatacttccacagag

M295 = EIF1AY STS20c, (507bp) **T** to **C**, at position 411,

(Group VIII). STS also contains M294 mutation

catggtccaagcaatttattttgTGAGTTCCCAAAATAATTTATACAGCAATGATTCATGTG
ACAATGTGAATAAATAGAAAAAGTCTTTGATAACTTTTAGATTTACTTTTAAA
GAATAATTTGTTTGTTTAACTTCTGTTGTATTCCTACCAGAAATGTTTACTCTG
ATATTAGTATTGAAGAAACCAGACAAATCTAATATATAACACAAATGGTCTT
GACTCAGATGTTAATGCTGTGAAAGAATGAAAAATCTGGGAATTACTTTAGC
TTAAAAGAGATTGATCGGTGCATATCCCTTTGTTAGGTTTTGGATTGGGGGAA
ATAGTTTTAGGTGGTACTAGGAAAAATTGGAATATGTTAGAAACTC
TATTTGTTAGTAATACCACATCAGGTAGTTTYATAAAATTACACTGATTAAAAG
TCTCTACTACTCAGATTTTTAATTAAAAATAAAAAACTTATTTTTGGCTGAGc
tctgtggaagtattagccagc

F 5'-3' = catggtccaagcaatttatttttg

R 5'-3' = gctggctaatacttccacagag

M296 = EIF1AY STS21=STS20d, (536 bp) C to T, at position 165,

(Group VIII)

F 5'-3' = gattgggggaaatagttttagg

R 5'-3' = cactatgtgtggactaagccag

M297 = EIF1AY STS24, (506 bp) **A to G**, at position 303, (Group VII)

TtggttggtctacgggactATCAGGTAAAAATAACATTTAAAGTTGTGGTATGTCTGTGT
TTAAGCAGTTGTTAATGTTTGGAAGGTAACTATACTAGCATCTTTGACCCATT
CCAGCCCAGGTTGCTTTCTCACCATTCTGCCTGCCATCATCATCATTATTAAGGG
CCAGTTGTATTTCAGACTATAGTATTTTTCAAATTTGACATAATTCTCACTGAT
AGTAAATGGTACATATATTTTTGTGGAAAGACATAAAGTTTTTAATTCTTTGT
TTTCATTGTTAATATATATGTGCAGTAAATRTTTTCTTGCAGGCTTGGGCAAGT
ACTGTAGACCATCTGTCCTCATCCATTTAAAGGCCAATGGTGTTTCAGGCATT
CAGCTAGGTATTTCAGACATTGTAGTTCCCAAATGCCGGTCTGTTAAATAGTA
TTGGTGCAGGCTGAATTTTCAGTGCTCTGAAGTCAAATTAGAAGATACATAGT
Tacgatgtttttcatggagca

F 5'-3' = ttggttggtctacgggact

R 5'-3' = tgctccatgaaaaacatcgt

M298 = EIFIA STS 27 (445 bp) G to A at position 230,

Group II

Original R 5'-3' = tgagatcctgcacagcaaga

M299 = EIF1AY STS29, (483 bp) **T** to **G**, at position 127,

Group I

TGTCCATTATATTCGAATTATCATTTTAACAAAAACTGGTTTGCACATTA CAGTTTGAAAAGTGTTGGTCTATTTCATactgccattgtgacagatca

F 5'-3' = cggacttggtctgtgcttttc

R 5'-3' = tgatctgtcacaatggcagt

M300 = EIF1AY STS31, (500 bp) **G** to **A** at position 153,

STS also contains M301, Group III

F 5'-3' = caggcaggtctactttcaatct

R 5'-3' = gtcaaacactgcaattcaaaac

M301 = EIFIA STS 31 (500 bp) A to C at position 340bp.

(Group III) STS also contains M300, a GroupVII marker

F 5'-3' = caggeaggtetacttteaatet

R 5'-3' = gtcaaacactgcaattcaaaac

M302 = EIFIA STS 32a (527bp) A to G at position 230

(Group VII)

GATATTTTGGTATAAGACGTTTTGAAAGTTATTTGTTTATTTCTAAGGATAAC AAAGCTGATGTAATTTTAAAGTacaatgcagatgaagctagaag

F 5'-3' = caaagtgctgggattacagg

R 5'-3' = cttctagcttcatctgcattgt

M303 = EIFIA STS 32b (527bp) G to C at position 352, (Group X)

CaaagtgctgggattacaggCGCGAGCCACCGTGCCTGGCCTAGAAAAGTGTATTACCT
TTTTAACATCATTATTCTTTACTCCATTTTTAGTTTTGAATTGCAGTGTTTGAC
CTTAAAAGTTTTATATTACAATTTTTTTAATTAGTCTTTTATTTTTTCCAAGAG
ACTTCTAATTAAAAGGGAATAGTAAATAAAAGCACTGTGCTTGCCTTTTGTGC
TTTTATTAAAGTGAAATCTCTACAATCTTTCCTAAGCTGTTAATCACTGTTTAC
TAATGAACATAAACCACTTCCTAATTATTCAGACTCAAGAATTTTTTTCTAGA
GGGTATTGGGGTAGGCAAAGAAAASCAGGAGGTTTGTAACAAACAGTATG
TGGGATTTTTTAGATGTGTTCAATTTGAAAGTAACTTGTGAAACAACTGGTG
ATATTTTGGTATAAGACGTTTTGAAAGTTATTTGTTTATTTCTAAGGATAACA
AAGCTGATGTAAATTTTAAAGTacaatgcagatgaagctagaag

F 5'-3' = caaagtgctgggattacagg

R 5'-3' = cttctagcttcatctgcattgt

M304 = EIFIA STS 32c (527bp) A to C at position 421

F 5'-3' = caaagtgctgggattacagg

R 5'-3' = cttctagettcatctgcattgt

M305 = EIFIA STS 33 (545 bp) **C** to **T** at position 331 (Group I)

AacttgtgaaacaactggtgatATTTTGGTATAAGACGTTTTGAAAGTTATTTGTTTATTTC
TAAGGATAACAAAGCTGATGTAATTTTAAAGTACAATGCAGATGAAGCTAGA
AGCCTGAAGGCATATGGCGAGCTTCCAGAACATGGTAAGATCAAAAATGATTT
TATCTCCTCATTATTTGATATTAATGTTTGTTGGTATTTAGGTGAAGGTATTTC
CGTAGAACTCTTGTTTTACATACTGTTTTAGTGTATACTTAAAAAATTTGTTATA
AGTAGTCTTGCCTATACTTCAGTTTACTTATGATACTTTGGAAAAGATATTAA
TAAYTGGAAATCTCTAATAAAAAACGTTATGAACTTGAAAGTAGAAGTCTCTA
ATAAAGAGATTATGAATTATGAAAGTTCCTTTAGTGACAACTTTATAAATTCA
TAAGCTCTGGATTTGTATATAAAGATCTGTCAAAGAAATACGTTTTTTATAGTG
TTTTTCTAAACAGTTCTCAAGACTGGCAGTTTTCATTTaagcagaggcaacaaatgtaat

F 5'-3' = aacttgtgaaacaactggtgat

R 5'-3' = attacattlgttgcctctgctt

M306 = EIFIA STS 34b (399 bp) C to A at position 231.

Group IX. STS also contains M284, a Group VI marker.

F 5'-3' = ggcagttttcatttaagcaga

R 5'-3' = agcgaaactttcagcacttc

M307 = EIFIA STS 35 (500 bp) G to A at position 282 (Group VI)

F 5'-3' = ttattggcatttcaggaagtg

R 5'-3' = gggtgaggcaggaaaatagc

M308 = EIFIA STS 37a (444 bp) **T to C** at position 70 (Group I)

F 5'-3' = aaactttacagtcctttgggata

R 5'-3' = tctcaagagagaaaaaatc

M309 = EIFIA STS 37b (444 bp) A to G at position 200

(Group II)

F 5'-3' = aaactttacagtcctttgggata

R 5'-3' = tctcaagagggaagaaaaatc

M310 = EIFIA STS 37c (444 bp) C to T at position 352

(Group III)

F 5'-3' = aaactttacagtcctttgggata

R 5'-3' = tctcaagagagaaaaaatc

M311 = EIFIA STS 39 (460 bp) **G to T** at position 304

(Group X)

F 5'-3' = cgagaacagcctaaccaaca

R 5'-3' = gggtgtgatagatgaagcagag

M312 = EIF1AY STS40a, A to T at position 49,

(Group VII)

gtttccagactgttcagaggagTAGAAGGATTTTTAAATTTATTTGTAWACATTCAAATAC TCACCAACAATATTGTACAATTTACAGTTTTTCTCTGCTTCATCTATCACACCC ATCCTTCTATTCATCTGATATTACACCTTATATTTTGGCACATTTCCAAACTAT

TACTTACACTTTGAGTTGAAGAAAATAAACTGAGTCCTTAATTGTATTGTATA
TATGCATTTATAAATTTTTACAACATAAAGTACTCTATATTTACAAAATTTTTT
AGTTTTTTTTTCTTTGGAATTGTTTCTGAGTAGTACTTAGTAACACTACTCTA
ATGTAATATAAATTTTAAAGTATACCCAAAAAGAAAATGAAAAGAGATGAA
AAATGCATTGTTCTTGTGATCCCAGGAAATCTGAGACAGGTCTCAGTTAATTT
acaaagttgattttgccaaagt

F 5'-3' = gtttccagactgttcagaggag

R 5'-3' = actttggcaaaatcaactttgt

M313 = EIFIA STS 40b Homopolymer 9T's to 10T's at position 288

For 5'-3' = gtttccagactgttcagaggag

Rev 5'-3' = actttggcaaaatcaactttgt

M314 = EIFIA STS 40c (623 bp) A to C at position 419. (Group VI)

F 5'-3' = gtttccagactgttcagagg

R 5'-3' = aaggetaacaagatgeeete

M315 = EIFIA STS 41 (512 bp) A to C at position 395 STS also contains M314 GttcttgtgatcccaggaaatCTGAGACAGGTCTCAGTTAATTTACAAAGTTGATTTTGCC AAAGTTGAGGACGCCCATGACACAGCCTCGGGAAGCCCTGAGGACATGT ACCCAAGGTGTTTGGGGCACAGCTTGGTTTACTACATCTTCAGGGAGACATG AGACATCAATCAATATATGTGAAAAGAACGTTGGTTCAGTTTGGAAAGGAG

GGCATCTTGTTAGCCTTTCTAAAGGAGGCAGTCAGCTATGCATCTAACTCAAT GAGCGAAAGGATAACTTTTGAATAGAATGGGAGGCCGGTTTGTCTTAAGCAG TTTCCACCTTGAGTTTTTCATAGTAATTTTGGGGGCCAAAGATATTTTCGTTTC ACATTCTAATATTTTCTTC**M**TGTACCTCCCTTTGGGGACCCTGAGCCAGAGGT TTTTTGGGGGATTAAACAGAATTGGCATTTACTTCATGTTGCAATAACCAAAA GCATAAATAttttgttgtagattaagggcaa

F 5'-3' = gttcttgtgatcccaggaaat

R 5'-3' = ttgcccttaatctacaacaaaa

M316 = EIFIA STS 42 (512 bp nominal) 5T's to 6T's at position 201 Group V

F 5'-3' = aattggcatttacttcatgttgc

R 5'-3' = catgtccttacttccttttgtg

M317 = EIFIA STS 44 (523 bp nominal) –2bp Deletion of GA at position 400 (Group VIII)

F 5'-3' = tggttctacagttgggattttg

R 5'-3' =ccttaataaccgaggcacaa

M318 = EIF1AY STS20d, T to C, at position 353 Group VI

Catggtccaagcaa:ttattttTGTGAGTTCCCAAAATAATTTATACAGCAATGATTCATGT GACAATGTGAATAAATAGAAAAAGTCTTTGATAACTTTTAGATTTACTTTTAA AGAATAATTTGTTTGTTTAACTTCTGTTGTATTCCTACCAGAAATGTTTACTCT GATATTAGTATTGAAGAAACCAGACAAATCTAATATATAACACAAATGGTCT TGACTCAGATGTTAATGCTGTGAAAGAATGAAAAATCTGGGAATTACTTTAG CTTAAAAGAGATTGATCGGTGCATATCCCTTCGTTAGGTTTTGGATTGGGGGA F 5'-3' = catggtccaagcaatttatttttg

Rev 5'-3' = gctggctaatacttccacagag

M319 = UTY1 exon 14b, T to A at position 124. Group VI

GtaaaactcagatatatacatccatgAAATATACACAGAAACTATAAATTAGCATTAATATC CTCTAAAATGATACTGTAGTAAAGAAAATATTCTCAAACTGTTGGTAAATTTA GAGAAAAWAAAAATATTATACATACTTGCTGCATTAAGACAAACTGACTTTC TAACTGTTCCAGCTGATGCTTCTGTGCTGGATTTAAATTATCTCTATTTGCTCG CAGTTGTTCCAAGTGCTAGAAGAAAAGAGATTAATATAATCAAAGTTTAATC TAAAATTTAAGACAATATAAGGCAACTCCTCACTAAAAAGACTACACAGAAC CTTTGCAGGATGAAAGACAGTGATTCCTAATGAACgttaagatagtgattctttttttt

F 5'-3' = gtaaaactcagatatatacatcccatg

Rev 5'-3': aaaaaaaaagaatcactatcttaacg

M320 = DBY STS08, (444 bp) T to G at position 60

Group VI

For 5'-3'=tgaggtggaatgtatcagtatacc

Rev 5'-3'=tgatttcaaggatttgttagtctt

M321 = DBY STS08, (444 bp) C to T at position 171 group VI

For 5'-3'=tgaggtggaatgtatcagtatacc

Rev 5'-3'=tgatttcaaggatttgttagtctt

Footnote:

STS sequences (one strand only) for polymorphic Y sequences.

Primer regions = lower case; Reverse compliment made to generate 5'-3' Reverse PCR primer sequence for complimentary strand.

IUB code defines polymorphic site

R = A or G (puRine)

Y = C or T (pYrimidine)

K = G or T (Keto)

M = A or C (aMino)

S = G or C (Strong-3H bonds)

W = A or T (Weak-2H bonds)

H = A, C or T

Markers M1, M29, M40, M46, M130, M167, M176, M177, M222, M236, M288 are unassigned in TABLE 1.